


**REVIEW**

# Consensus guidelines for the diagnosis and management of isolated sulfite oxidase deficiency and molybdenum cofactor deficiencies

Bernd C. Schwahn<sup>1,2</sup>  | Francjan van Spronsen<sup>3</sup> | Albert Misko<sup>4</sup> |  
 Julija Pavaine<sup>5,6</sup> | Victoria Holmes<sup>7</sup> | Ronen Spiegel<sup>8</sup> | Guenter Schwarz<sup>9</sup> |  
 Flora Wong<sup>10</sup> | Alistair Horman<sup>11</sup> | James Pitt<sup>12</sup> | Jörn Oliver Sass<sup>13</sup> |  
 Charlotte Lubout<sup>3</sup>

<sup>1</sup>Division of Evolution & Genomic Sciences, Faculty of Biology, Medicine and Health, University of Manchester, Manchester, UK

<sup>2</sup>Manchester Centre for Genomic Medicine, Saint Mary's Hospital, Manchester University Hospitals NHS Foundation Trust, Manchester Academic Health Science Centre, Manchester, UK

<sup>3</sup>Department of Metabolic Diseases, Beatrix Children's, University Medical Center (UMC) Groningen, Groningen, Netherlands

<sup>4</sup>Massachusetts General Hospital Department of Neurology, Boston, Massachusetts, USA

<sup>5</sup>Department of Paediatric Radiology, Royal Manchester Children's Hospital, Manchester University Hospitals NHS Foundation Trust, University of Manchester, Manchester, UK

<sup>6</sup>Division of Informatics, Imaging & Data Sciences, School of Health Sciences, Faculty of Biology, Medicine and Health, University of Manchester, Manchester Academic Health Science Centre, Manchester, UK

<sup>7</sup>Willink Unit, Manchester Centre for Genomic Medicine, St Mary's Hospital, Manchester University NHS Foundation Trust, Manchester, UK

<sup>8</sup>Department of Pediatrics Unit B, Emek Medical Center, Afula, Israel

<sup>9</sup>Center for Molecular Medicine Cologne, Köln, Germany

<sup>10</sup>Monash Children's Hospital/Hudson Institute of Medical Research/Monash University, Melbourne, Victoria, Australia

<sup>11</sup>Great Ormond Street Hospital, London, UK

<sup>12</sup>Victorian Clinical Genetics Services, The Royal Children's Hospital, Parkville, Victoria, Australia

<sup>13</sup>RG Inborn Error of Metabolism, Department of Natural Sciences & Institute for Functional Gene Analytics (IFGA), Bonn-Rhein-Sieg University of Applied Sciences, Rheinbach, Germany

**Correspondence**

Bernd C. Schwahn, Division of Evolution & Genomic Sciences, Faculty of Biology, Medicine and Health, University of Manchester, Manchester, UK.  
 Email: [bernd.schwahn@mft.nhs.uk](mailto:bernd.schwahn@mft.nhs.uk)

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**Abstract**

Sulfite intoxication is the hallmark of four ultrarare disorders that are caused by impaired sulfite oxidase activity due to genetic defects in the synthesis of the molybdenum cofactor or of the apoenzyme sulfite oxidase. Delays on the diagnosis of these disorders are common and have been caused by their unspecific presentation of acute neonatal encephalopathy with high early mortality, followed by the evolution of dystonic cerebral palsy and also by the lack of easily available and reliable diagnostic tests. There is significant variation in survival and in the quality of symptomatic management of affected children. One of the four disorders, molybdenum cofactor deficiency type A (MoCD-A) has recently become amenable to causal treatment with synthetic cPMP

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(fosdenopterin). The evidence base for the rational use of cPMP is very limited. This prompted the formulation of these clinical guidelines to facilitate diagnosis and support the management of patients. The guidelines were developed by experts in diagnosis and treatment of sulfite intoxication disorders. It reflects expert consensus opinion and evidence from a systematic literature search.

#### KEYWORDS

consensus guidelines, cPMP, fosdenopterin, molybdenum cofactor deficiency, sulfite oxidase deficiency

## 1 | INTRODUCTION

Sulfite in the human organism mainly derives from catabolism of the sulfur amino acids methionine and cysteine and is oxidized to non-toxic sulfate by sulfite oxidase (EC 1.8.3.1.). Extremely low or absent sulfite oxidase activity leads to excessive accumulation of sulfite.<sup>1-3</sup> Sulfite is highly reactive. It exerts direct toxic effects on mitochondrial energy metabolism<sup>4-7</sup> and cleaves disulfide bonds including that of cystine to form S-sulfocysteine (SSC)<sup>8</sup> which has specific neuro-excitatory properties on NMDA receptors.<sup>9-11</sup>

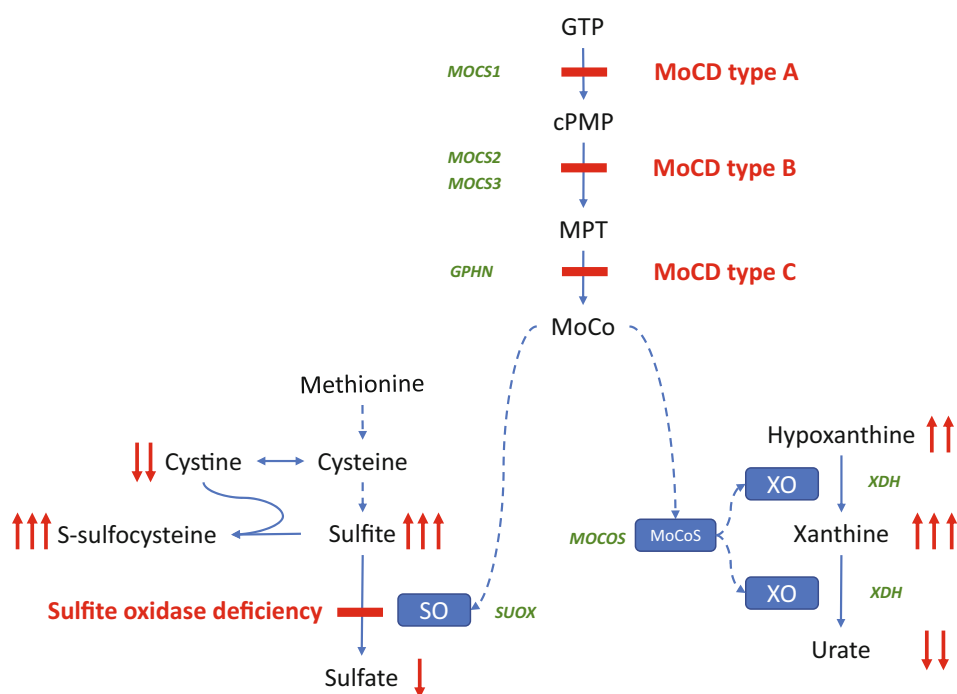
Sulfite intoxication disorders are ultrarare with a birth prevalence of less than <1:100 000 in most populations.<sup>12</sup> Primary, isolated sulfite oxidase deficiency (ISOD, OMIM #272300) is caused by biallelic pathogenic variants in the *SUOX* gene. Secondary, combined sulfite oxidase deficiency can arise from genetic disorders that disrupt the de novo synthesis of molybdenum cofactor (MoCo) which is required for 4 oxidoreductases in humans, including sulfite oxidase, xanthine oxidase (EC 1.17.3.2), aldehyde oxidase

(EC 1.2.3.1), and mitochondrial amidoxime reducing complex (EC 1.16.98.B1). MoCo is synthesized by a 3-step biosynthetic pathway that involves the products of 4 genes, *MOCS1*, *MOCS2*, *MOCS3*, and *GPHN* (see Figure 1).<sup>2,13</sup> Biallelic pathogenic variants in *MOCS1* result in MoCD type A (MoCD-A, OMIM #252150), also known as cyclic pyranopterin monophosphate (cPMP) synthase deficiency, which has been found in around 60% of published patients.<sup>14</sup> Most other patients have been diagnosed with MoCD type B (OMIM #252160) caused by *MOCS2* or, rarely, *MOCS3* mutations. Only single cases of MoCD type C (caused by *GPHN* mutations, OMIM #615501) are currently known.<sup>13-15</sup>

Why are these guidelines required?

Numerous case reports have demonstrated a deficit in recognizing and a delay in diagnosing sulfite intoxication disorders, as well as significant uncertainty and variation in the management of affected children. Treatment of severely affected children is supportive and symptom control is difficult and often unsatisfying.

**FIGURE 1** Molybdenum cofactor synthesis and MoCo-dependent pathways. Red bars denote distinct disorders and their level of biochemical disruption. Gene symbols are depicted in green. Blue boxes symbolize enzymes, blue solid arrows indicate enzymatic reactions and blue dotted lines multiple or non-enzymatic steps. Metabolites are written in black and red arrows indicate changes in metabolite concentrations. cPMP, cyclic pyranopterin pyrophosphate; GTP, guanosin triphosphate; MoCo, molybdenum cofactor; MoCoS, molybdenum cofactor sulfurase; MPT, molybdopterin; SO, sulfite oxidase; XO, xanthine oxidase.



The advent of a novel treatment for MoCD type A, namely substitution with cPMP, has created heightened interest in sulfite intoxication disorders. Since 2008, cPMP has been available in clinical trials and in compassionate use programs for a small number of individual patients.<sup>16</sup> Treatment with synthetic cPMP (fosdenopterin) has been granted market authorization for the treatment of MoCD type A by FDA in February 2021, by the Israeli Ministry of Health in July 2022 and by EMA in September 2022. The evidence base for the rational use of cPMP is very limited. These clinical guidelines were developed to facilitate diagnosis and support the management of patients.

## 2 | METHODS

### 2.1 | Guideline development

These guidelines were developed by experts with documented experience in the diagnosis and treatment of sulfite intoxication disorders. It reflects expert consensus opinion and evidence from a systematic literature search. The guideline development group was constituted following an international workshop on the diagnosis and management of MoCD and ISOD in May 2021, sponsored by the British Inherited Metabolic Disease Group (BIMDG) and supported by an unconditional educational grant from Origin Biosciences Inc, which served to identify key questions. Further development of the guideline was supported by a guideline development grant from the Society for the Study of Inborn Errors of Metabolism (SSIEM).

Clinical experts were largely comprised of specialists from centers that had participated in previous clinical trials of cPMP substitution in MoCD type A, from 2009 to 2022. Laboratory scientists with established expertise in diagnosing sulfite intoxication disorders were co-opted. Four guideline development group meetings were held between January 2022 and March 2023. The draft guidelines were submitted for review to international clinical and laboratory experts with published experience in diagnosing and managing children with sulfite intoxication disorders. Some guideline development group members and external experts were unable to comment on particular statements where those fell outside of their specific area of expertise. This was noted for each statement. The final draft was shared with parents of affected children and representatives of a patient advocacy organization (Metabolic Support UK) inviting further comments. All comments were considered and incorporated as far as possible.

### 2.2 | Competing interests

All members of the guideline development group were required to report potential conflicts of interest. AM, JPa,

VH, AH, JPi, and JOS declare they have no conflict of interest. BS, RS, CL, FvS, FW were investigators on one or more clinical trials sponsored by either Colbourne Pharmaceuticals GmbH, Alexion Inc, or Origin Biosciences Inc. BS reports personal fees from Origin Biosciences Inc for taking part in an advisory board meeting. RS reports personal fees from Origin Biosciences Inc and Sentynl Inc, for lectures and for taking part in advisory board meetings. GS is co-inventor on a patent on the use of cPMP in the treatment of MoCD type A and CEO of Colbourne Pharmaceuticals GmbH. He reports royalties and personal fees from Origin Biosciences Inc and Sentynl Inc. These conflicts of interest were carefully considered while formulating recommendations about the use of cPMP.

### 2.3 | Systematic literature review

PubMed (<http://www.ncbi.nlm.nih.gov/pubmed>) and the Cochrane library (<https://www.cochranelibrary.com>) were searched in January 2022 using the following search terms:

[sulfite oxidase deficiency OR sulfite oxidase deficiency OR molybdenum cofactor deficiency OR SUOX OR MOCS1 OR MOCS2 OR MOCS3 OR GPHN OR Gephyrin] AND [human]

Three hundred fifty-six peer-reviewed publications were identified, from an initial search result of 700, and a further 69 more recent articles were added manually by GDG members to be incorporated into the final draft where relevant. Publications were collected and made available to GDG members in a literature database using the software Zotero (<https://www.zotero.org>).

Articles were filtered according to disease (Molybdenum Cofactor Deficiency, Sulfite Oxidase Deficiency, Gephyrin Deficiency) and type of publication (Review, Single Case Report, Case Series, Clinical Trial, Laboratory Study only) and further tagged to facilitate their evaluation, using the terms: Clinical Presentation, Genetic Testing, [Genetic Therapy], Biochemical Testing, [Immunohistochemistry Testing], Brain Imaging, Dietary Treatment, Drug Treatment, cPMP, [Newborn Screening], [New Diagnosis Method], [Fetal Autopsy], [Animal Testing].

### 2.4 | Grading and strength of recommendations

Guideline development group members used the Grading of Recommendation Assessment, Development and Evaluation (GRADE) approach<sup>17,18</sup> to assess the quality of the evidence and determine the strength of recommendation for each statement (see Table 1).

**TABLE 1** Evidence levels and definitions modified after Grading of Recommendation Assessment, Development and Evaluation (GRADE).<sup>17–19</sup>

Level of evidence	Definition	Examples
(A) High quality	Further research is very unlikely to change our confidence in the estimate of effect	Experimental controlled trial (with or without randomization); Prospective cohort study; Systematic review demonstrating high level of consistency and low risk of bias
(B) Moderate quality	Further research is likely to have a relevant impact on our confidence in the estimate of effect and may change the estimate	Case-control study with consistent large size of effect and low risk of bias. Residual confounding would reduce the effect estimate
(C) Low quality	Further research is very likely to have an important impact on our confidence in the estimate of effect and is likely to change the estimate	Case series or case reports with possible confounders identified; serious inconsistency; serious risk of bias
(D) Very low quality	Any estimate of effect is very uncertain	Expert opinion; case reports with very serious inconsistency or imprecision or high risk of bias

Due to the rarity of sulfite intoxication disorders, the available evidence is mostly anecdotal, with a complete absence of controlled clinical trials. However, the publication of two prospective cohort studies, four large case series and four systematic reviews of the clinical and laboratory manifestations allowed upgrading the level of evidence for some statements, owing to a consistently large effect size and low risk of bias.<sup>19</sup> A few good practice statements were included to highlight certain aspects or care and provide contextual background information. Those statements represent expert consensus and were not assessed using GRADE, as systematic research and evaluation of their underlying evidence was beyond the remit of these guidelines.

The strength of statements and recommendations relied on the level of agreement from experts. A statement or recommendation received a strong endorsement when there was at least 90% agreement (completely agree or partly agree) of all responding experts and conditional recommendations were made if at least 50% of experts agreed.

## 2.5 | Consensus procedure

A draft guideline was developed and submitted for review in three phases. First, the draft was circulated for review within the guideline development group. Second, the draft together with a link to an online survey on all statements and recommendations was sent to guideline development group members and to a further international group of 29 clinical and laboratory science experts on the subject. Based on a total of 22 qualified responses, revisions were made. The final draft was sent out for a second consultation before submission for publication.

## 3 | DIAGNOSIS

### 3.1 | Clinical manifestation

The clinical presentation of MoCD and ISOD has been evaluated in four recent systematic reviews.<sup>20–23</sup> In addition, Spiegel et al.<sup>24</sup> reported data from a retrospective and prospective natural history study including 58 genetically confirmed patients with MoCD that had not previously been published. Table 2 lists the main sources of evidence from these 5 publications and other large case series.

Children with ISOD or MoCD are usually born at term with birth measurements in the normal range and manifest with severe encephalopathy and seizures after a variable symptom-free interval during the neonatal period, with a majority presenting within 24–72 h after birth. Symptoms of encephalopathy in neonates can be subdued and the disease may not be recognized until later in infancy, especially in cases with antenatal onset of encephalopathy. Between 12% and 18% of children present after the neonatal period.<sup>21,23,24</sup> Most patients present as “typical” “early-onset,” “neonatal,” and “severe” cases. A minority presents as “atypical,” “late-onset” or “post-neonatal,” and “mild” or “attenuated” cases. These terms have been used inconsistently in the literature. Importantly, late neonatal or post-neonatal clinical manifestation does not preclude the development of severe neurological sequelae during infancy. The age at manifestation does not generally differ between ISOD and MoCD subtypes, although there is a trend to a higher proportion of severe and earlier presentations in reported cases of ISOD.<sup>21,23</sup> This may partly be caused by an ascertainment and reporting bias.

#### Statement 1

Children with sulfite intoxication disorders present with characteristic but mostly unspecific symptoms. The acute presentation of ISOD and MoCD is clinically indistinguishable. Two, partly overlapping, clinical syndromes

**TABLE 2** Overview of available evidence from cohort studies, observational studies, larger case series and systematic reviews.

Type of evidence	Population studied	Reference
Cohort studies	MoCD (N = 16)	Schwahn et al. 2015 <sup>25</sup>
	MoCD (N = 21 out of 58)	Spiegel et al. 2022 <sup>24</sup>
Cross-sectional studies	MoCD (N = 58)	Spiegel et al. 2022 <sup>24</sup>
Case series	MoCD (N = 8)	Vijayakumar et al. 2011 <sup>26</sup>
	MoCD (N = 12)	Bayram et al. 2013 <sup>27</sup>
	MoCD (N = 6), ISOD (N = 3)	Zaki et al. 2016 <sup>28</sup>
Systematic reviews	MoCD (N = 82, 1980–2013)	Mechler et al. 2015 <sup>20</sup>
	ISOD (N = 47, 1967–2016)	Claerhout et al. 2018 <sup>21</sup>
	MoCD-B (N = 35)	Arıcan et al. 2019 <sup>22</sup>
	MoCD (N = 94) and ISOD (N = 52) incorporating references 20–22, 28	Misko et al. 2020 <sup>23</sup>

Note: The number of included patients and time period covered by systematic reviews is given in parentheses.

have been observed that can be differentiated by age at clinical onset of symptoms and by severity of neurological sequelae in the chronic phase.

We recommend using the term “typical” for individuals presenting antenatally or up to 4 weeks after birth with acute encephalopathy or seizures and severe global abnormalities in neuroimaging, with resultant dystonic spastic quadriplegia and severe developmental impairment during infancy. The term “atypical” should be used for children presenting first disease-related symptoms after the neonatal period, with or without a later acute encephalopathic episode, and with neurological symptoms of variable severity, including dystonia, motor developmental delay, speech delay, and stroke-like episodes, corresponding to focal abnormalities in brain imaging.

*Level of evidence: A*

*Strength of recommendation: Strong*

*Expert opinion: 90% agree, 5% partly agree, 5% disagree (2/22 unable to comment)*

**Q1.** Are there characteristic clinical signs that should prompt diagnostic investigations?

### Typical manifestation

Fetal seizures may be noted during late pregnancy by mothers as “increased hiccupping” and some children are compromised at birth, suggesting perinatal asphyxia. The typical neonatal presentation is characterized by acute encephalopathy with sudden or insidious onset and rapidly progressive symptoms, including lethargy, feeding difficulties, irritability, hyperekplexia, apnea and seizures, as well as truncal hypotonia and variable appendicular tone. Entering the acute encephalopathic phase infants appear distressed and can have an altered, high-pitched cry.

Surviving infants become more alert after 1–2 weeks. Seizures persist or recur after a period of apparent improvement. Over the following months, children develop secondary microcephaly and display severe intellectual impairment, epilepsy as well as visual impairment, whereas hearing is preserved. The most prominent symptom is a severe dyskinetic quadriplegic movement disorder, characterized by prominent dystonia and moderate spasticity, with anarthria, un-co-ordinated swallow, laryngeal stridor, opisthotonic crises as well as frequent myoclonic spasms and hyperekplexia.<sup>29</sup> Seizures may initially be controlled but then become pharmaco-resistant.<sup>2</sup> Ectopia lentis with uni- or bilateral lens subluxation without directional predilection<sup>30</sup> occurs in a high proportion of infants and young children but is not always present. In MoCD, xanthine nephrolithiasis can occur at any age.

During the first few months of life infants often share facial features of prominent cheeks, broad nasal bridge, widely spaced eyes, elongated palpebral fissures, and a long philtrum<sup>2,23,31</sup> (see Figure 2).

### Statement 2

The typical manifestation of sulfite intoxication disorders is characteristic but not specific to ISOD and MoCD. Their presentation partly overlaps with that of disorders of mitochondrial energy metabolism and can be mistaken for other more common causes of neonatal encephalopathy due to perinatal hypoxia, neonatal stroke, or encephalitis.

A diagnosis of ISOD or MoCD should be considered in all neonates presenting with encephalopathy and seizures, especially if there is no history of peripartum complications and in those with diffuse brain injury without evidence of perinatal hypoxia or infection.

*Level of evidence: A*

*Strength of recommendation: Strong*

*Expert opinion: 100% agree (2/22 unable to comment)*

### Atypical presentation

Atypical cases of ISOD or MoCD can present with variable neurological symptoms during childhood and the absence of neonatal encephalopathy or seizures does

**FIGURE 2** Facial features of molybdenum cofactor deficiency in an affected child at age 3 weeks and at age 2.5 years. Note prominent cheeks, broad nasal bridge, widely spaced eyes, elongated palpebral fissures, long philtrum and facial expression of distress at neonatal age; small neurocranium in childhood due to microcephaly.



not exclude a diagnosis.<sup>15,23,32–40</sup> Attenuated presentations include ataxia, dystonia or choreoathetosis on a background of normal or delayed development and children can experience acute or gradually progressive encephalopathy with decreased consciousness and seizures. Clinical presentations similar to Leigh syndrome and with developmental regression have been described<sup>41</sup> Ectopia lentis can occur after infancy. A mild phenotype of MoCD type B due to a specific *MOCS2* gene variant manifesting with developmental delay, hypouricemia and xanthinuria has recently been described in a Roma population.<sup>42</sup> Acute deterioration of an attenuated disease can be triggered by febrile illness or by mechanical head trauma.<sup>43</sup>

#### Statement 3

The atypical presentation of sulfite intoxication disorders includes a variety of neurological symptoms, commonly involving the extrapyramidal motor system.

The differential diagnosis ISOD or MoCD should be considered in children presenting with acute onset of dystonia, seizures or encephalopathy, especially on the background of a previous dyskinetic disorder or of motor or global developmental delay.

*Level of evidence: B*

*Strength of recommendation: Conditional*

*Expert opinion: 85% agree, 15% partly agree (2/22 unable to comment)*

Antenatal presentation.

See under 3.3 Neuroimaging.

### 3.2 | Laboratory diagnosis

Sulfite accumulation is the primary consequence of impaired sulfite oxidase activity due to ISOD or MoCD. In MoCD, the additionally impaired function of xanthine oxidoreductase also leads to accumulation of

hypoxanthine and xanthine and to a lack of urate.<sup>2,44</sup> Very few other ultrarare metabolic disorders are known that can lead to a moderate accumulation of sulfite,<sup>45</sup> or to hypouricemia with accumulation of hypoxanthine and xanthine.<sup>46</sup>

#### Sulfite

Sulfite in body fluids can be directly measured. Due to the reactivity of sulfite, testing for urinary sulfite requires fresh or at least directly frozen urine. As sulfite ( $\text{SO}_3^{2-}$ ) is in equilibrium with hydrogen sulfite ( $\text{HSO}_3^-$ ) and sulfur dioxide ( $\text{SO}_2$ ) its quantitation may in addition be affected by a low urinary pH value  $<6$ .<sup>47</sup> Quantitative measurements of sulfite in plasma or urine have been reported but are not routinely available in clinical practice. Usually, a dip-stick urine test is used for semiquantitative assessment of sulfite and any presence of sulfite is considered a positive test result. Notably, commercially available test strips are usually designed for water or food analysis and are not certified as medical devices. Both false negative<sup>48,49</sup> and false positive sulfite tests are well known to occur, with sulfhydryl-group containing drugs like mucolytic 2-mercaptoethanesulfonate and several antibiotics known as causes of the latter.<sup>50,51</sup> While a lack of urinary sulfate as a consequence of sulfite oxidase deficiency has been postulated,<sup>52,53</sup> this is not a consistent finding.<sup>45,49</sup>

#### Statement 4

A positive urine sulfite dip-stick test in a neonate or infant with a typical presentation of ISOD or MoCD should raise suspicion of a sulfite intoxication disorder. Due to its unreliability this test is not sufficient to confirm or rule out the diagnosis.

*Level of evidence: C*

*Strength of recommendation: Conditional*

*Expert opinion: 100% agree (1/22 unable to comment)*

S-sulfocysteine in urine, plasma and cerebrospinal fluid (CSF)

S-sulfocysteine (SSC) is formed by nucleophilic cleavage of cystine disulfide bonds by sulfite.<sup>8</sup> SSC is chemically more stable than sulfite. It has been measured in urine and plasma using conventional ion-exchange column chromatography amino acid analysis<sup>52,54</sup> and its presence strongly suggests a sulfite intoxication disorder. SSC can be quantified using tandem mass-spectrometry or high-performance liquid chromatography (HPLC) with pre-column derivatization with *o*-phthalaldehyde (OPA).<sup>55</sup> It is present in small amounts in urine of healthy individuals and increased 5–50 fold in ISOD or MoCD.<sup>23,55,56</sup> There is no overlap of urinary SSC concentrations between healthy and affected individuals. While there is interindividual variation in concentrations, and poor discrimination of patients with typical and atypical manifestation,<sup>23,24</sup> the intraindividual range of SSC remains relatively stable over time.<sup>24</sup>

Quantification of SSC in urine is available in specialized clinical diagnostic laboratories. Elevated SSC can also be measured in extracts from dried blood spots<sup>57</sup> and in plasma and CSF samples, but there is limited availability of these tests in clinical practice.

#### Statement 5

S-sulfocysteine is currently the most reliable and valid laboratory marker of sulfite accumulation and should be used for the biochemical confirmation of a suspected sulfite intoxication disorder.

*Level of evidence: A*

*Strength of recommendation: Strong*

*Expert opinion: 90% agree, 10% partly agree (2/22 unable to comment)*

#### Taurine in urine or plasma

Elevated urinary taurine levels have frequently been reported in patients with ISOD or MoCD and taurine and hypotaurine were consistently elevated over 20-fold in urines of 9 patients with MoCD compared with adult controls.<sup>58</sup> Upregulation of the taurine biosynthesis pathway from cysteine sulfinic acid via hypotaurine to taurine has been postulated as a possible mechanism.<sup>58</sup> Taurine is included in routine quantitative amino acid profiles. Its diagnostic value is however limited since an elevation of plasma taurine is a frequent observation in healthy newborns and infants<sup>59</sup> and taurine in plasma or urine is not consistently increased in ISOD or MoCD [authors' observation and reference 60].

#### Statement 6

Plasma and urinary taurine concentrations may be increased in ISOD and MoCD, but this finding is neither sufficiently sensitive nor specific to be of diagnostic use.

*Level of evidence: C*

*Strength of recommendation: Strong*

*Expert opinion: 94% agree, 6% partly agree (4/22 unable to comment)*

#### Thiosulfate

Accumulating sulfite will partly be converted to chemically stable thiosulfate. Urinary thiosulfate has been found to be elevated in sulfite oxidase deficiency<sup>54,61</sup> although the test has not been widely established in diagnostic laboratories. Notably, depending on methodology, antibiotic interference can result in false-positive<sup>62,63</sup> and false-negative<sup>64</sup> results. However, recent methodological advances are likely to promote the use of this parameter for laboratory diagnostics as well as for therapeutic monitoring.<sup>65</sup> Measurement of thiosulfate, in conjunction with SSC, as part of multiplex urine screening by mass spectrometry, may assist in the diagnosis of ISOD and MoCD and circumvent issues of sulfite instability.

#### Statement 7

Thiosulfate is a biomarker of sulfite accumulation. There is currently limited access to reliable measurement of thiosulfate in clinical diagnostic laboratories and insufficient published evidence to recommend its routine use for the diagnosis of ISOD or MoCD.

*Level of evidence: C*

*Strength of recommendation: Conditional*

*Expert opinion: 80% agree, 13% partly agree, 7% disagree (7/22 unable to comment)*

#### Plasma total homocysteine

Plasma total homocysteine is a compound measurement comprised of a small proportion of free reduced homocysteine and a larger pool present as free homocysteine, free mixed disulfide or protein-bound disulfide. Accumulation of sulfite leads to the formation of stable S-sulfohomocysteine which escapes detection in commercial assays and causes an apparent decrease of the plasma total homocysteine pool.<sup>45,66,67</sup> Plasma total homocysteine quantification is widely available in routine clinical chemistry. A decreased concentration, close to<sup>68</sup> or below the quantitation limit,<sup>60,69</sup> is a rare finding that strongly points to excessive sulfite accumulation and a sulfite intoxication disorder. However, total homocysteine is not always severely decreased in children with attenuated sulfite intoxication disorders and the lower end of the reference interval is not well defined.

#### Statement 8

Plasma total homocysteine should be measured in infants with intractable seizures or abnormal movements of uncertain cause because it is widely available and can provide reliable indirect evidence of sulfite accumulation. A plasma total homocysteine concentration below the reference interval in a child with suggestive features should prompt additional diagnostic tests for ISOD or MoCD.

*Level of evidence: C*

*Strength of recommendation: Strong*

*Expert opinion: 90% agree, 10% partly agree (2/22 unable to comment)*

### Plasma cystine

Similar to L-homocysteine, L-cysteine is present in plasma in small amounts as free cysteine and as a larger pool of free or protein-bound disulfides. Accumulation of sulfite leads to cleavage of disulfides and the formation of S-sulfocysteine.<sup>45</sup> The free disulfide cystine is often quantified as part of routine plasma amino acid analysis and it is decreased in sulfite intoxication disorders.<sup>60,67,68,70</sup> Unfortunately, free plasma cystine is particularly prone to pre-analytical cleavage, and any delay between sampling and centrifugation and freezing causes artificially low levels.<sup>71</sup> Similarly, cystine quantitation can be unreliable when using some amine derivatization methods, for example with 6-aminoquinolyl-N-hydroxysuccinimidyl carbamate, that is employed in some modern tandem MS amino acid assays.

#### Statement 9

A severely decreased or undetectable plasma cystine can be a marker of sulfite accumulation. Due to the poor stability of plasma cystine and analytical issues, there is a high risk of falsely low results and large variability between laboratories. A plasma cystine concentration in the average or high reference interval, however, renders a diagnosis of sulfite intoxication disorder unlikely.

*Level of evidence: C*

*Strength of recommendation: Conditional*

*Expert opinion: 81% agree, 19% partly agree (6/22 unable to comment)*

#### Xanthine and Hypoxanthine in urine or plasma

In molybdenum cofactor deficiency, but not in isolated sulfite oxidase deficiency, purine abnormalities represent additional primary biomarkers.<sup>51</sup> Lack of the molybdenum cofactor inactivates xanthine oxidase, which leads to a significant accumulation of hypoxanthine and xanthine, while urate in plasma and urine is decreased or even undetectable.<sup>2,24</sup>

Hypoxanthine and xanthine are sensitive markers of impaired xanthine oxidoreductase activity and have consistently been abnormal, even in attenuated cases of MoCD.<sup>24</sup> Measurements are however only available in specialized metabolic laboratories.

#### Statement 10

An increased concentration of xanthine and/or hypoxanthine without a concomitant increase in urate is a reliable and sensitive marker of reduced xanthine oxidase activity and can point to the diagnosis of MoCD.

*Level of evidence: A*

*Strength of recommendation: Conditional*

*Expert opinion: 76% agree, 24% partly agree (5/22 unable to comment)*

#### Urate in plasma and urine

In typical MoCD, urate concentrations in plasma and urine are found to be decreased below the

reference interval after the first few days of life. Urate can, however, temporarily remain in the normal range during the first few postnatal days, owing to delayed clearance of plasma urate originating from transplacental maternal supply. Urate concentrations can remain low-normal in atypical cases of MoCD with attenuated biochemical presentation.<sup>24</sup> Moderate hypouricemia can also be caused by renal tubular dysfunction, medication, total parenteral nutrition or neoplastms.<sup>46</sup> A very low or absent plasma urate can indicate a renal tubular defect of urate re-absorption and, if it occurs in conjunction with very high concentrations of xanthine and hypoxanthine, may indicate an isolated defect in xanthine oxidase, due to mutations in the *XDH* gene, or other ultrarare disorders such as MoCo sulfurase deficiency or purine nucleoside phosphorylase deficiency.<sup>46,72</sup> However, these conditions lack the abnormalities in sulfur metabolism which are always present in MoCD.<sup>73</sup>

#### Statement 11

Urate is a widely and readily available parameter in routine clinical chemistry. A severely decreased urate concentration, in conjunction with signs of sulfite accumulation strongly points to a diagnosis of MoCD whereas a normal concentration in body fluids does not always rule out MoCD.

Plasma urate should be measured in every child suspected of a sulfite intoxication disorder. In this clinical context, a decreased urate should be followed up by analysis of purines in plasma or urine.

*Level of evidence: A*

*Strength of recommendation: Strong*

*Expert opinion: 95% agree, 5% partly agree (2/22 unable to comment)*

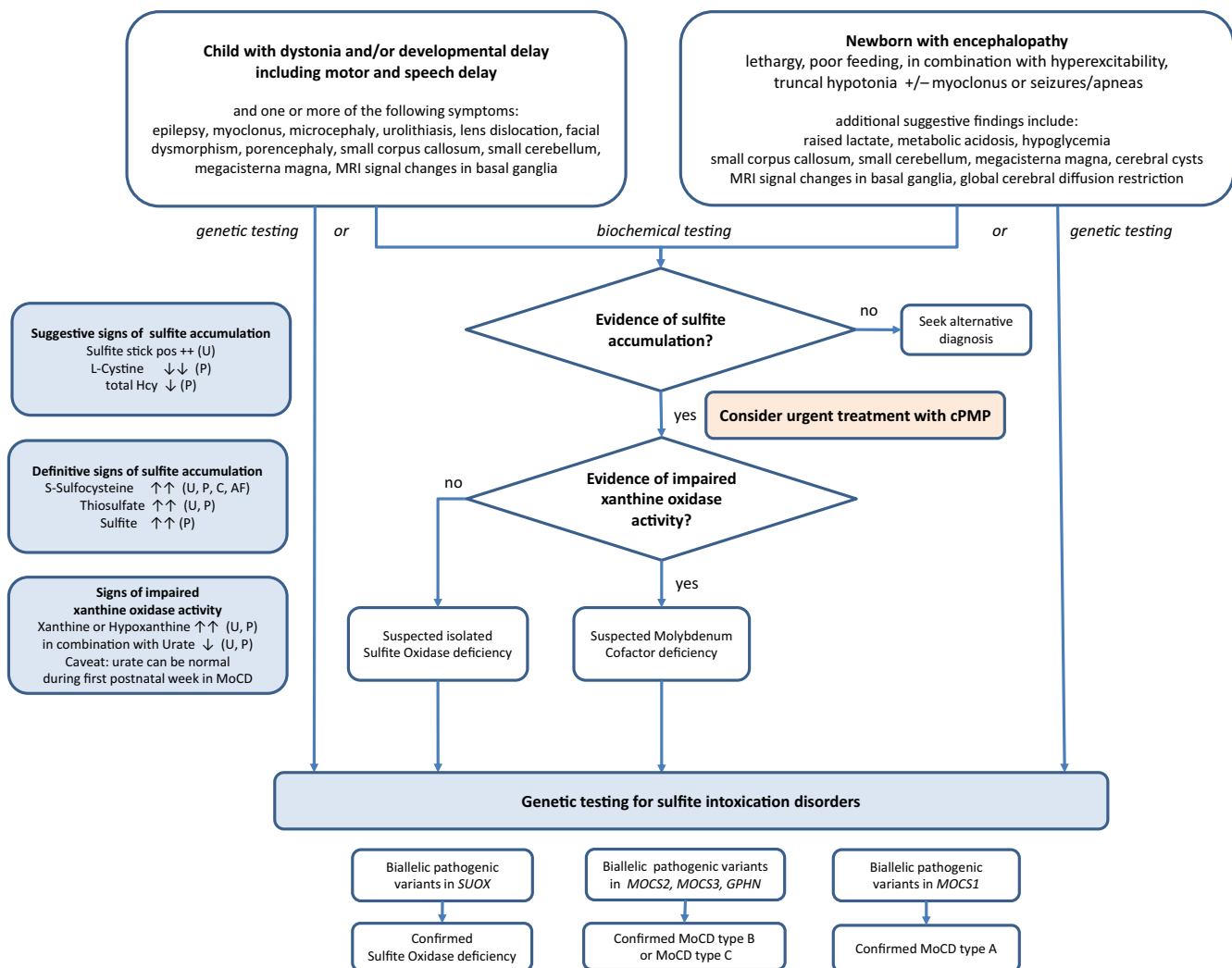
#### Alpha amino adipic semialdehyde

Alpha amino adipic semialdehyde (a-AASA) is moderately increased in urine of patients with ISOD and MoCD but this finding is not specific for sulfite intoxication disorders.<sup>74,75</sup> Increased AASA can point to a diagnosis of ISOD or MoCD.<sup>76</sup>

#### Experimental biomarkers

Increased S-sulfonation of plasma proteins such as transthyretin can be used as a marker of excessive sulfite accumulation.<sup>67,77</sup> Compound Z is the oxidation product of cPMP and is decreased in MoCD type A.<sup>3,65</sup> This parameter is however only available in research laboratories. Lack of urothione, a degradation product of MoCo, in urine of patients with molybdenum cofactor deficiency<sup>78</sup> provides a basis for the use of this biomarker to diagnose MoCD. However, since it has been available in research settings only, it did not gain a major role. A recent study casts doubt on the specificity of urothione as a biomarker since polymorphisms in the *TPMT* gene,





**FIGURE 3** Diagnostic flowchart for suspected sulfite intoxication disorders. Availability of biochemical and genetic testing is highly variable between countries. The most appropriate strategy to rapidly establish a diagnosis should be chosen according to local availability.

encoding thiopurine S-methyltransferase, may also result in extremely low urinary urothione concentrations.<sup>79</sup>

**Q2.** Are there diagnostic tests available that allow a timely and reliable diagnosis?

**Statement 12**

In the presence of clinical features suggestive of a sulfite intoxication disorder, simple and readily available biochemical tests such as sulfite urine dip stick, plasma urate, plasma amino acid analysis or plasma total homocysteine can raise suspicion of ISOD or MoCD but all these tests are prone to produce false positive or false negative results. Any such suggestive evidence of sulfite accumulation should however prompt further urgent investigation for ISOD or MoCD.

A definitive diagnosis can be achieved by measuring biochemical markers including SSC and urinary purines

or by genetic testing. Molecular genetic testing can reveal biallelic unequivocally pathogenic variants but it has limitations due to variable availability of rapid testing facilities and pertinent issues with variant detection and interpretation. Whether to use primary genetic or biochemical testing will depend on local availability and reporting times. We recommend a testing strategy as outlined in Figure 3.

*Level of evidence: B*

*Strength of recommendation: Strong*

*Expert opinion: 100% agree*

*Reference intervals and quality assurance for biomarkers:*

In the laboratory diagnostics of rare diseases, the selection of commercially available calibrators and control materials is limited. Reference intervals for biomarkers of sulfite accumulation are variable between laboratories and not always robustly defined. Table 3 lists

**TABLE 3** Key biomarkers for isolated sulfite oxidase deficiency and molybdenum cofactor deficiencies and typical findings in affected children.

Parameter	Matrix	Reference population	Isolated sulfite oxidase deficiency	Molybdenum cofactor deficiencies
Sulfite	Urine	Negative	Positive	Positive
Total homocysteine	Plasma	~5–12 $\mu\text{mol/L}$	<5 $\mu\text{mol/L}$	<5 $\mu\text{mol/L}$
S-sulfocysteine	Urine	<20 $\mu\text{mol/mmol creatinine}$ <sup>55,57</sup>	2–15 fold greater than ULN	2–15 fold greater than ULN <sup>24</sup>
	Plasma	<3 $\mu\text{mol/L}$ <sup>55</sup>	2–10 fold greater than ULN	2–10 fold greater than ULN <sup>24</sup>
Urate	Plasma/urine	Within age-appropriate reference interval	Within age-appropriate reference interval	Variably low Plasma urate can be normal during the first few postnatal days and will become undetectable in typical cases during the first postnatal week
Hypoxanthine	Urine	Within reference interval	Within reference interval	2–10 fold greater than ULN <sup>53</sup>
	Plasma	Within reference interval	Within reference interval	1–10 fold greater than ULN <sup>53</sup>
Xanthine	Urine	Within reference interval	Within reference interval	2–50 fold greater than ULN <sup>24,53</sup>
	Plasma	Within reference interval	Within reference interval	1–10 fold greater than ULN <sup>24</sup>

Note: ULN is upper limit of normal (reference interval).

key biomarkers for isolated sulfite oxidase deficiency and molybdenum cofactor deficiencies with exemplified reference intervals and typical abnormal values in patients. Rarely, patients with atypical clinical presentation may present with less abnormal findings.

Sulfite standards in water (traceable to standards of the National Institute of Standards and Technology, NIST) and originally intended for calibration of quantitative analysis by ion chromatography is provided by suppliers such as VWR International Ltd (Lutterworth, UK). The scheme “Quantitative Amino Acids (serum)” by ERNDIM ([www.erndim.org](http://www.erndim.org)) allows external quality control of S-sulfocysteine, taurine and cystine levels. Homocysteine is covered by the ERNDIM schemes “Special Assays in Serum” and “Special Assays in dried blood spots.” L-cystine and S-sulfocysteine analysis in urine can be validated using the ERNDIM material for “Special Assays in Urine.” External quality control for homocysteine in serum is also available from Referenzinstitut für Bioanalytik ([www.rfb.bio](http://www.rfb.bio)), while Instand e.V. ([www.instand-ev.de](http://www.instand-ev.de)) includes cysteine-homocysteine-disulfide, cystine, and taurine into its scheme for amino acid analysis. Quantitative analyses of hypoxanthine and xanthine can be controlled using the ERNDIM reference material “Purines and Pyrimidines (urine),” while its concentration of uric acid does not vary and is part of the matrix. However, external control material for uric acid is, for example, provided by Instand e.V. ([www.instand-ev.de](http://www.instand-ev.de)), Referenzinstitut für Bioanalytik [www.rfb.bio](http://www.rfb.bio), or UK-NEQAS.

Good practice statement 13

Each laboratory should regularly review its external quality assurance requirements against available programs.

### 3.3 | Neuroimaging and EEG

**Q3.** Are there characteristic results from neuroimaging that should prompt diagnostic investigations?

Fetal imaging

Multiple case reports provide evidence of antenatal manifestations of severe MoCD. Prenatal brain imaging of fetuses with ISOD or MoCD can reveal cerebral cysts, brain atrophy, poor gyration, poorly developed corpus callosum or a megacisterna magna from as early as 21 weeks of gestation.<sup>70,75,80–83</sup> Fetal seizures may be noted by the mother as “increased hiccupping” in some cases. Prenatal magnetic resonance imaging (MRI) in two affected fetuses showed a megacisterna magna and slightly smaller cerebellum from 32 weeks gestational age (GA). From 36 weeks GA mild ventriculomegaly and a slight increase in signal intensity of the cerebral white matter was noted on T2 weighted imaging, suggesting white matter edema.<sup>84</sup> Of particular concern are reports of prenatal multicystic encephalomalacia that were detected at 35 weeks GA<sup>75</sup> and 14 h after birth,<sup>85</sup> respectively, and which suggest that severe sulfite-related prenatal brain injury can occur prior to birth.

## Statement 14

Structural developmental brain abnormalities, progressive cerebral white matter edema, and multicystic encephalomalacia are typical postnatal finding but can sometimes be found in antenatal brain imaging of fetuses with ISOD or MoCD. ISOD or MoCD should be considered if fetal ultrasound or fetal MR imaging reveals multicystic lesions in subcortical regions or basal ganglia or if a megacisterna magna is present in combination with additional findings, specifically, if increased T2 signal intensity in hemispheric white matter or cortical diffusion restriction is found during the third trimester of pregnancy on fetal MRI.

*Level of evidence: B*

*Strength of recommendation: Strong*

*Expert opinion: 89% agree, 11% partly agree (3/22 unable to comment)*

Neuroimaging during the phase of acute neonatal encephalopathy

Brain MRI during the acute phase of neonatal sulfite-related encephalopathy is characterized by widespread strikingly severe diffusion restriction involving the cortex at the depths of sulci,<sup>86</sup> T2 hyperintensity in the cerebral cortex and in subcortical white matter with gyral swelling, as well as T2 hyperintensity in basal ganglia with an increased lactate peak on MR spectroscopy. Ultrasound or MR imaging can reveal subcortical cysts at birth and striking diffusion restriction can be seen as early as 4–24 h after birth in symptomatic infants.<sup>82,87</sup> Additional structural abnormalities as described under the prenatal presentation are frequently seen in both ISOD and MoCD.<sup>21–23,26,39,81,85,86,88,89</sup> The brain MRI appearance has similarities with that of severe global hypoxic-ischemic brain injury (HIE).<sup>90,91</sup> Within 2 weeks after the onset of acute encephalopathy, imaging reveals brain volume loss with marked ulegyria and emerging cystic encephalomalacia. Diffusion restriction can persist beyond the acute phase.<sup>92,93</sup>

## Statement 15

Acute sulfite intoxication causes a brain MRI appearance of generalized acute vasogenic and cytotoxic edema with early neuronal necrosis that mimics that of severe global hypoxic brain injury. In contrast to HIE, typical anatomical abnormalities can often be found and true diffusion restriction can persist after the first week of acute encephalopathy.

*Level of evidence: A*

*Strength of recommendation: Strong*

*Expert opinion: 94% agree, 6% partly agree (5/22 unable to comment).*

Neuroimaging in typical cases after the neonatal period

After recovery from acute neonatal encephalopathy, infants and children develop a typical brain MRI

appearance including diffuse brain atrophy, cavitory subcortical encephalomalacia and persistent hypotrophy of the corpus callosum and of cerebellar structures<sup>23,26</sup> (see Figure 4). Infants can develop subdural effusions and bleeds, secondary to severe brain atrophy.

## Statement 16

Brain imaging after the neonatal period mimics that of children who suffered from severe perinatal hypoxic brain injury. Persistent abnormalities of the corpus callosum and cerebellum are frequent findings.

*Level of evidence: A*

*Strength of recommendation: Strong*

*Expert opinion: 89% agree, 11% partly agree (3/22 unable to comment)*

Neuroimaging in patients with atypical presentation

In patients with post-neonatal and attenuated manifestations, brain MRI can reveal isolated T2 hyperintense lesions and diffusion restriction in globi pallidi<sup>94,95</sup> and in the cerebellum<sup>26,28</sup> as well as abnormalities including mega cisterna magna, cerebellar hypotrophy and/or cystic lesions in the globi pallidi.<sup>35,94,95</sup> Rarely, infants without a history of acute severe encephalopathy and presenting after the neonatal period have been found to have the typical brain MRI appearance of generalized cystic encephalomalacia, likely indicating prenatal onset of disease.

## Statement 17

Lesions in the globi pallidi with diffusion restriction, a mega cisterna magna and cerebellar hypotrophy in patients presenting with movement disorder, dystonia, hemiplegia and/or seizures should prompt investigations to rule out a sulfite intoxication disorder.

*Level of evidence: C*

*Strength of recommendation: Conditional*

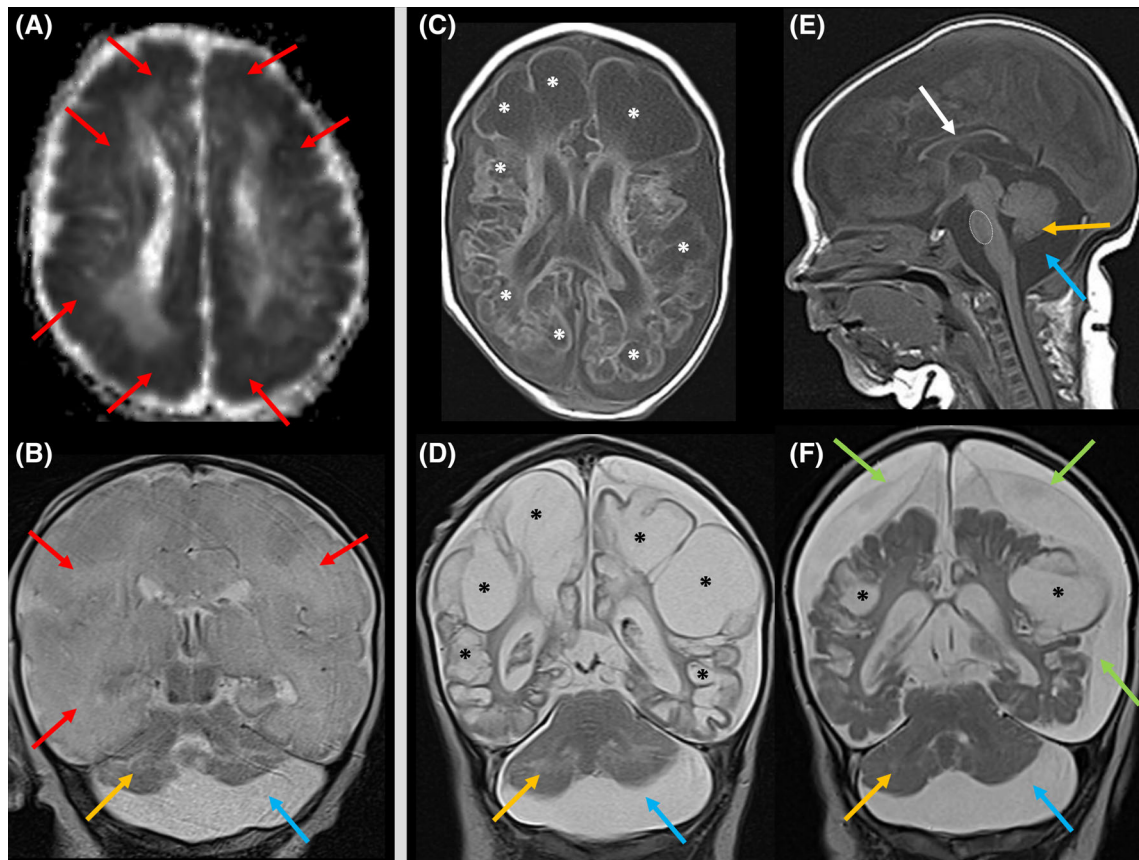
*Expert opinion: 83% agree, 17% partly agree (4/22 unable to comment)*

Diagnostic value of EEG in neonates with sulfite intoxication

Severe encephalopathy in infants with sulfite intoxication disorders during the acute manifestation, is reflected in a burst suppression pattern in the EEG.<sup>80</sup> A burst-suppression pattern is not specific and can also be the result of anticonvulsant treatment. It is not predictive of the extent of permanent injury. One group suggested delta-beta complexes might be a specific diagnostic marker for sulfite toxicity.<sup>96</sup> Current evidence does not suggest that EEG aids in the diagnosis of sulfite intoxication disorders.

### 3.4 | Diagnosis through generalized newborn screening programs

**Q4.** Should universal newborn screening for sulfite intoxication disorders be considered?



**FIGURE 4** Neuroimaging in a typical case of MoCD in the acute and chronic stages of the disease. Left column, (A, B)—acute stage at 6th day of life; middle and right columns, chronic stages (C–E) at 24th day of life and (F) at 48th day of life. (A) (axial ADC map) shows hyposignal in the entire cerebral cortex and subcortical white matter (red arrows) indicative for striking diffusion restriction due to cytotoxic oedema in the acute stage. (B) (coronal T2WI) demonstrates hypersignal in the cerebral white matter (red arrows), diffuse gyral swelling and sulcal effacement reflecting brain oedema in the acute stage. (C) (axial T1WI) and (D, F) (coronal T2WI) depict extensive bilateral cerebral cystic encephalomalacia (asterisks) in the chronic stage. (F) (coronal T2WI) shows collapse of most of the encephalomalacic cysts, bilateral subdural effusions (green arrows) reflecting severe brain atrophy in the chronic stage. (E) (sagittal midline T1WI) demonstrates severe hypoplasia of corpus callosum (white arrow), small pons with reduced pontine protuberance (dotted circle), hypoplastic cerebellar vermis (orange arrow), and mega cisterna magna (blue arrow). Lower row, (B, D, F) (coronal T2WI) denote evolving brain change from cerebral oedema in the acute stage to cystic encephalomalacia and subsequent atrophy in the chronic stage. Note persistent hypotrophy of the cerebellar hemispheres (orange arrows) and mega cisterna magna (blue arrow).

Untargeted whole genome or whole exome sequencing has helped to identify ISOD and MoCD in symptomatic children.<sup>97,98</sup> Selective biochemical screening for disorders of purine metabolism in children with neurological disease can also yield an unexpected diagnosis.<sup>73,99</sup> Since a majority of children manifests with the typical neonatal presentation, they will likely experience a severe encephalopathic crisis before the newborn screening result becomes available, which limits the benefit of the intervention. Universal newborn screening for sulfite intoxication disorders would however contribute to an earlier diagnosis in some cases, especially those with atypical presentation where a diagnostic delay is more likely.

Statement 18

Biochemical or genetic universal NBS would shorten the diagnostic pathway for children with typical and atypical disease manifestation and an earlier diagnosis would impact at least on the management of attenuated cases, allowing earlier consideration of specific treatment options. At present, there is insufficient data about the birth prevalence of sulfite intoxication disorders, especially of cases with atypical presentation, and about the effectiveness of general newborn screening to allow an evaluation of the benefit of this intervention.

*Level of evidence: C*

*Strength of recommendation: Conditional*

*Expert opinion: 86% agree, 14% partly agree (0/22 unable to comment)*

## 4 | DISEASE COURSE, TREATMENT, AND ONGOING SURVEILLANCE

### 4.1 | Disease course, typical sequelae, and complications

**Q5.** Which typical sequelae and complications are caused by MoCD/ISOD and need to be considered during follow-up?

The pattern and prevalence of chronic manifestations of MoCD and ISOD have recently been systematically reviewed (Table 2).

After the acute encephalopathic period, children with typical manifestation of ISOD or MoCD suffer from axial hypotonia with or without appendicular spasticity. They invariably show developmental arrest, cognitive impairment, generalized seizures, myoclonus, cortical visual impairment with or without nystagmus, and secondary microcephaly. Ectopia lentis and enophthalmos can develop after infancy. In one systematic evaluation, the most common MoCD sequelae were limb hypertonia (84.5%), spastic quadriplegia (56.9%) or diplegia (24.1%), severe global developmental delay (81.0%), truncal hypotonia (74.1%), dysmorphic facial features (67.2%), and acquired microcephaly (63.8%).<sup>24</sup> A particular problem in infants and young children are frequent dystonic crises with opisthotonus that can be triggered by minor stimuli. Recurrent and frequent vomiting is a common feature and may represent autonomous neuropathy and intestinal dysmotility. Pyloric stenosis has been found over proportionately frequently in the acute encephalopathic and post-acute phase of disease manifestation in ISOD<sup>100</sup> and in MoCD.<sup>92,101,102</sup> Urinary xanthine stones have been reported in infants and children with MoCD.<sup>103,104</sup>

Some or all of these symptoms can be found in children with attenuated disease, however often with later onset, in milder form and with a higher proportion of extrapyramidal movement disorders, including choreoathetosis, dystonia, and ataxia.<sup>21,23,24</sup> Seizures are less common in attenuated cases.<sup>28,38</sup>

#### Statement 19

Typical clinical signs and symptoms have been consistently reported in the chronic, post encephalopathic phase of children with severe ISOD and MoCD. Disease sequelae affect the central and peripheral nervous system and the integrity of connective tissues. Signs are more variable in attenuated cases. A typical presentation with neonatal severe encephalopathy invariably leads to profound disability. Children who present with atypical manifestations can still experience acute or slowly

progressive deterioration later in life, with severe neurological sequelae.<sup>24</sup>

*Level of evidence: A*

*Strength of recommendation: Strong*

*Expert opinion: 94% agree, 6% partly agree (4/22 unable to comment)*

### 4.2 | Genotype phenotype correlation and prognosis

**Q6.** Is there a reliable genotype–phenotype correlation and can a genotype predict the prognosis?

Hinderhofer et al.<sup>105</sup> assessed the genotype–phenotype correlation of 40 published cases of MoCD. The genotype was classified as severe or mild based on in-silico prediction and was correlated with survival data. Patients with a predicted severe genotype showed a median survival of 15 months and had a lower probability of survival compared to patients with predicted mild genotypes who were all alive at last reported follow-up.

Claerhout et al.<sup>21</sup> assessed 31 variant *SUOX* alleles from published cases. They concluded that most alleles occurred in the homozygous state and were associated with severe disease. Only two variants were associated with late-onset disease, namely NM\_000456.2 variant c.182T>C p.Leu61Pro in the transit peptide<sup>35</sup> and variant c.427C>A p.His142Asn in the heme binding domain.<sup>94,106</sup>

Consistent with a high consanguinity rate, most patients in a study of 58 patients with MoCD had homozygous pathogenic variants.<sup>24</sup> Generally, mutations were private or were shared by individuals from the same ethnic group, apart from three variants that were found in a few individuals from different ethnic groups, in particular c.217C>T in *MOCS1* which was found in three ethnic groups. The low allele frequency of variants in *MOCS1* did not suggest the presence of multiple mutational hot spots. Several mutations were associated with post-neonatal onset and/or longer survival, including c.1338delG, c.1165+6T>C, c.377G>A, c.949C>T, c.394C>T, c.1000dupT, and c.1102+1G>A in *MOCS1* (using reference sequence NM\_001358530.2) and c.3G>A, and c.57A>T in *MOCS2* (using reference sequence NM\_004531.5). Age at onset of symptoms and long-term outcomes were however variable in children with such genotypes.

Misko et al.<sup>23</sup> evaluated data of 146 published patients of whom defects in *MOCS1*, *MOCS2*, *MOCS3*, *GPHN* and *SUOX* were reported in 62 patients. The authors established a good correlation between the presence of

deleterious variants and severe clinical outcomes whereas variants with less severe effects on gene products were associated with variable outcomes. Further comprehensive reviews of variants in *MOCS1*,<sup>107</sup> *MOCS2* and *MOCS3*<sup>108</sup> demonstrate that patients with typical manifestation have genetic variants that abolish the function of their gene product whereas patients with atypical presentations are presumed to have some residual function. However, disease severity and time until onset of encephalopathy can vary to some extent between children and even between siblings with the same genotype.<sup>26,33,95</sup>

#### Statement 20

Phenotypical variability in ISOD and MoCD is largely but not exclusively explained by the genotype. Only a few genotypes are known to be consistently associated with an atypical manifestation and milder symptoms in both ISOD and MoCD. These genotypes are associated with residual function of the gene product.

*Level of evidence: B*

*Strength of recommendation: Strong*

*Expert opinion: 88% agree, 12% partly agree (5/22 unable to comment)*

### 4.3 | Disease management

**Q7.** Are there specific requirements for treatment and monitoring of patients with MoCD and ISOD?

#### Antenatal and perinatal management

Obstetric ultrasonography or fetal MRI can reveal structural abnormalities from as early as 21 weeks of gestation<sup>70</sup> and progressive white matter edema or even brain necrosis during late pregnancy. Multiple reports of prenatal cerebral manifestations of sulfite intoxication disorders suggest that treatment prior to birth may be beneficial and have prompted consideration of premature induction of labor<sup>84,109</sup> for children with MoCD type A to reduce antenatal exposure to rising sulfite during late pregnancy and to enable earlier specific treatment with cPMP. Premature delivery is associated with its own risks and it is not known at present whether the premature brain is equally or perhaps more vulnerable to exposure to a postnatal surge of sulfite and SSC, compared with that of a term neonate.

#### Statement 21

At present, there is insufficient evidence to generally recommend premature delivery for infants with MoCD type A. Premature delivery of fetuses affected with MoCD-A should be carefully considered in every individual case.

*Level of evidence: C*

*Strength of recommendation: Conditional*

*Expert opinion: 84% agree, 16% partly agree (2/22 unable to comment)*

#### Statement 22

We recommend using prenatal cerebral imaging for fetuses that are at risk or known to be affected with MoCD type A. Serial fetal MRI scans from the second trimester of gestation will be particularly informative to identify progressive changes. Early identification of brain abnormalities will inform prognostic expectations and the decision whether to deliver affected children early. Because of the possibility of immediate postnatal onset of seizures and encephalopathy it is recommended to plan delivery in an obstetric unit with access to adequate neonatal critical care facilities and with immediate availability of cPMP for neonates affected with MoCD type A.

*Level of evidence: C*

*Strength of recommendation: Strong*

*Expert opinion: 100% agree (5/22 unable to comment)*

Management of the acute encephalopathic phase  
Neuroprotective agents and cerebral hypothermia

The disease course and brain imaging findings suggest that the postnatal rise in sulfite and related metabolites triggers a cascade of molecular events involving excitotoxicity, failure of mitochondrial oxidative phosphorylation, oxidative stress, and inflammatory factors leading to further secondary energy failure and extensive cortical and deep gray structures neuronal injury, in analogy to postulated mechanisms in hypoxic ischemic brain injury.<sup>110</sup>

There is evidence from animal and in vitro experimentation for a direct impairment of mitochondrial energy metabolism by sulfite<sup>4-7</sup> and for a strong excitotoxic effect of SSC on NMDA receptors<sup>10</sup> leading to neuronal apoptosis and necrosis. The NMDA receptor antagonist dextromethorphan was used at a dose of 12.5 mg/kg in a 3-year-old boy with MoCD and pharmaco-resistant epilepsy with good short-term effect.<sup>111</sup> A newborn with early severe ISOD was treated at the age of 3 weeks with dextromethorphan without positive effect.<sup>47</sup> Memantine was used in two neonates with MoCD with no discernible protective effect.<sup>92,112</sup>

Full-term infants with moderate to severe HIE are now routinely treated with hypothermia, ideally started within 6 h after birth, but the degree of neuroprotection remains incomplete. Treatment with hypothermia does not improve functional outcomes in infants with severe HIE and in premature infants.<sup>113</sup> There is no published evidence to suggest that cooling has been effective in preventing the typical severe sequelae of ISOD or MoCD.

#### Statement 23

NMDA antagonists have been neuroprotective in vitro and in vivo in an animal model of sulfite toxicity. There is limited experience in humans and currently no

sufficient evidence to recommend the regular use of NMDA antagonists in the acute phase of sulfite-related encephalopathy.

There is no clinical evidence of a benefit of cerebral cooling in ISOD or MoCD.

*Level of evidence: C*

*Strength of recommendation: Conditional*

*Expert opinion: 94% agree, 6% partly agree (6/22 unable to comment)*

Management of seizures and myoclonus

Pharmaco-resistant epilepsy is a common problem in children with a typical presentation of ISOD or MoCD. Anticonvulsive treatment is provided according to usual neuropediatric practice and often requires polymedication. No specific medication has been found to be particularly effective. Phenobarbital and midazolam are commonly used in neonates. Generalized seizures in older children are often treated with levetiracetam as a first choice, which may also be useful to treat myoclonus.<sup>114</sup> Clonazepam has been suggested as treatment of choice for cortical-subcortical myoclonus, with sodium valproate<sup>114</sup> as alternative. However, one group has cautioned against the use of valproate in sulfite intoxication disorders due to its interference with mitochondrial energy metabolism.<sup>23</sup>

Good practice statement 24

Anticonvulsive treatment for children with ISOD or MoCD should follow general recommendations. Theoretical concerns regarding the mitochondrial toxicity of sodium valproate should be considered when choosing the most appropriate medication.

Management of dystonia

Dystonia is a prominent symptom in the chronic phase of typical ISOD or MoCD and can be classified as a secondary-static dystonia.<sup>115,116</sup> Dystonia arises secondary to the sulfite-related postnatal neuronal brain injury to the basal ganglia and thalamus. It is clinically characterized by manifestation after the acute encephalopathic phase, usually within 3 months after birth. The dystonia is generalized and non-progressive, it often involves pharyngeal and laryngeal muscles with the distinct symptom of in- and expiratory stridor. Muscle contractions may appear persistent<sup>117</sup> but are usually absent in children at rest. Hypertonia can be triggered by minimal stimuli. Progression to a status dystonicus has been described, even as primary manifesting symptom in children with an atypical presentation.<sup>36,95</sup>

Dystonia management in children is not standardized and there is no disease-specific management of dystonia in ISOD or MoCD. Most commonly used drugs in the chronic management are baclofen, trihexyphenidyl and diazepam, which have however limited efficacy.<sup>118</sup> Gabapentin and clonidine<sup>119</sup> are also increasingly used. Due to

the minor contribution of spasticity and the paroxysmal nature of dystonia in ISOD and MoCD, one of the most effective interventions is to reduce triggering stimuli and reduce precipitants such as pain, infection, gastrointestinal discomfort due to gastro-esophageal reflux or constipation.

Providing good nutrition and hydration on the background of pharyngo-laryngeal dystonia and gastro-esophageal reflux and intestinal dysmotility usually requires tube feeding via gastrostomy or jejunostomy to reduce the risk of aspiration and pneumonia. Parents/carers will require adequate training and support with managing feeding and care at home.

Supportive care is important and includes analgesia and relief for respiratory distress or hypoxemia due to pharyngeal or laryngeal spasm or truncal dystonia. Parent feedback has highlighted the importance of calming measures and stress reduction as well as the utility of neurophysiotherapy to minimize distress. Sleep terminates dystonia, and sedation is often required to reduce or terminate distressing persistent dystonia after calming measures have failed. Sedation can be achieved with chloral hydrate or clonidine as well as cautious use of benzodiazepines.<sup>120</sup>

Good practice statement 25

Symptomatic management of dystonia is an important element of supportive care for children with ISOD or MoCD. Reducing stimuli that can trigger dystonia is often a very effective intervention. Intermittent sedation can be required to terminate dystonic crises.

Palliative care and end-of-life management

Complications from immobility, seizures and dystonia associated with severe ISOD or MoCD lead to a significant disease burden and, often, premature death. The median survival age in MoCD has been calculated as 3.0 years<sup>20</sup> and 4.23 years<sup>24</sup> for cohorts of patients with MoCD and mostly typical manifestation and as 2.5 years for a large cohort of children with ISOD or MoCD with typical manifestation.<sup>23</sup> Most children affected with ISOD will die prior to the age of 10 years.<sup>21</sup> Hospital admissions are frequently required to manage complications including respiratory infections. Infants and young children often appear severely distressed by discomfort or pain due to uncontrollable seizures and dystonic episodes.

Caring for affected children is very demanding and parents and siblings will usually require support from respite care or palliative care teams. There is a particular role for support groups and for patient organizations for rare diseases as a source of information and peer support.

Good practice statement 26

Access to physiotherapy, occupational therapy, respite care and to palliative care are important elements of health care support. The multidisciplinary nature of

care for a child with a typical presentation of a sulfite intoxication disorder requires good communication and co-ordination between health professionals, ideally led by specialists for neurodisability or palliative care. Families should always be directed toward relevant patient support organizations. Owing to a limited life expectancy, advance care planning should be considered early on.

#### 4.4 | Ongoing surveillance for specific complications

Lens dislocation can occur in ISOD or MoCD after infancy. This can lead to raised intra-ocular pressure and clinical symptoms of acute glaucoma.<sup>121</sup>

Gross accumulation of xanthine and hypoxanthine is observed in MoCD. Xanthine concentrations in urine easily exceed solubility thresholds.<sup>122</sup> Xanthine precipitates in urine can be found at any age and can lead to nephrolithiasis and acute urinary tract obstruction.<sup>103</sup> Nephrolithiasis is found to occur in around 40% of patients with isolated xanthine oxidase deficiency<sup>46,72,123</sup> and would be expected at the same frequency in MoCD. Good hydration and a low purine diet have been recommended. Both measures may be helpful<sup>70</sup> but are not generally sufficient to prevent urolithiasis.<sup>122</sup>

##### Statement 27

Regular ophthalmological follow up is recommended for all children affected with ISOD and MoCD to monitor for the manifestation of lens dislocation and potential intraocular hypertension.

For children with MoCD a renal ultrasound is recommended once yearly and as required in episodes of unexplained pain or distress to detect potential urinary tract infection and obstruction due to xanthine stones.

These recommendations do not apply to children with MoCD-A on cPMP supplementation.

*Level of evidence: C*

*Strength of recommendation: Strong*

*Expert opinion: 93% agree, 7% disagree (7/22 unable to comment)*

#### 4.5 | Specific treatment options

**Q8.** Is treatment with cPMP safe and effective in MoCD-A?

Is treatment with cPMP safe?

Daily doses of up to 980 µg/kg of cPMP as a free base have been used long-term in a small number of children without adverse drug reactions. Treatment emergent adverse events during clinical trials were reported

frequently and related to intercurrent illnesses and complications associated with central venous access (site infection, septicemia, or catheter blockage).<sup>16,124</sup> Animal studies have identified a potential risk of phototoxicity,<sup>16,124</sup> which has not unequivocally been observed in patients so far.

##### Statement 28

Substitution with cPMP in currently used doses is not associated with adverse drug reactions. Daily intravenous administration using implanted central venous catheters carries a risk of complications. Protection from avoidable skin UV exposure is recommended. Adequate care support should be provided to families of children who require daily intravenous infusions.

*Level of evidence: B*

*Strength of recommendation: Strong*

*Expert opinion: 94% agree, 6% partly agree (6/22 unable to comment)*

Can cPMP substitution correct the biochemical abnormalities in MoCD and does cPMP substitution require biochemical and clinical monitoring?

A biochemical response to dosing with cPMP has been consistently observed in all patients with MoCD type A. The pharmacodynamic biomarkers S-sulfocysteine, xanthine, hypoxanthine and urate respond within 24–48 h and return to normal or near-normal concentrations within 1 week.<sup>16,25,65,80,125</sup> No biochemical effect has been observed in patients with MoCD type B upon cPMP substitution.<sup>25</sup> Since the first treatment in 2008, no decrease in biochemical efficacy over time was observed, suggesting no requirement for frequent biochemical monitoring once a child is reliably established on treatment.<sup>25,126</sup>

##### Statement 29

We recommended regular biochemical monitoring of biomarkers during the first 2 weeks of cPMP substitution to document the response to treatment. The choice of biomarkers will vary depending on local availability. Once a response has been established, further, less frequent biochemical monitoring may be considered if there should be a clinical or regulatory requirement to document a sustained treatment effect and adherence to treatment.

*Level of evidence: A*

*Strength of recommendation: Strong*

*Expert opinion: 95% agree, 5% partly agree (3/22 unable to comment)*

Is there a dose–response relationship?

The initial cohort of neonates with MoCD-A was treated with a daily dose of intravenous recombinant cPMP starting with 80 µg/kg and increasing to 240 µg/kg after 3 months.<sup>25,65,80</sup>

Pharmacokinetic and pharmacodynamic studies in children with typical MoCD-A were undertaken with



daily doses ranging from 240 to 1300 µg/kg of cPMP monobromide dihydrate, which is equivalent to 180–980 µg/kg of the free base.<sup>127</sup> No pharmacodynamic data have been published to demonstrate a dose–response relationship or the benefits of higher doses.<sup>16</sup> The starting dose for the licensed preparation fosdenopterin (as free base) has been determined by the license holder as 400 µg/kg in pre-term infants and 550 µg/kg in term infants, administered once daily as slow intravenous infusion. A dose of 900 µg/kg once daily is suggested after the first 2 months of treatment and for any child that is over 1 year old at the start of treatment.<sup>124</sup> No experimentation has been undertaken to explore alternative dosing frequencies or modes of administration.

#### Statement 30

There is insufficient evidence to determine the optimum dose or dosing interval for cPMP substitution treatment.

*Level of evidence: C*

*Strength of recommendation: Conditional*

*Expert opinion: 69% agree, 31% partly agree (9/22 unable to comment)*

Does cPMP substitution improve clinical outcomes?

The overall survival of a cohort of children with MoCD-A treated with cPMP substitution was improved over a comparable cohort of untreated children.<sup>16,24,25,124,127</sup>

The long-term neurological outcome of children with MoCD-A treated with cPMP has been variable, depending on the extent of irreversible brain injury prior to treatment. A small number of neonates with MoCD-A that were treated with cPMP substitution prior to showing signs of severe acute sulfite encephalopathy have not developed cystic encephalomalacia and severe cerebral palsy, as opposed to their untreated affected siblings who displayed the typical phenotype of MoCD.<sup>25</sup> Their long-term psychomotor development has been much improved compared with untreated siblings or other untreated children with the same genotype. However, mild neurological symptoms such as speech delay, mild learning difficulties and central hypotonia have been observed despite treatment. Even very early postnatal treatment of two neonates, within a few hours after birth, who presented with seizures but no other signs of encephalopathy, could not prevent the development of mild to moderate neurological sequelae.<sup>80,84</sup>

Infants presenting with severely decreased level of consciousness and severe global diffusion restriction on brain MRI at the time of initiation of treatment have so far invariably developed typical severe clinical sequelae of MoCD, despite cPMP substitution.<sup>16,25</sup>

#### Statement 31

Substitution of cPMP in MoCD type A does improve long-term survival. The functional neurological outcome

depends on the timing of the intervention in relation to cerebral disease progress. Even very early postnatal treatment has been associated with some neurological sequelae. Treatment with cPMP that is started after onset of severe global diffusion restriction in brain MRI, corresponding to widespread neuronal necrosis, cannot prevent the manifestation of typical severe neurological sequelae. cPMP substitution should be urgently considered for any child with suspected acute sulfite intoxication until MoCD type A can be safely ruled out (see Figure 5).

*Level of evidence: B*

*Strength of recommendation: Strong*

*Expert opinion: 94% agree, 6% partly agree (4/22 unable to comment)*

#### Statement 32

A re-evaluation of treatment goals should be undertaken once the extent of irreversible brain injury is known. Discontinuation of cPMP treatment should be considered if the burden of treatment outweighs the achievable benefit.

*Level of evidence: C*

*Strength of recommendation: Strong*

*Expert opinion: 100% agree (2/22 unable to comment)*

### Q9. Is dietary modification of sulfur or molybdenum intake effective in MoCD/ISOD?

Does dietary sulfur restriction improve biochemical or neurological symptoms?

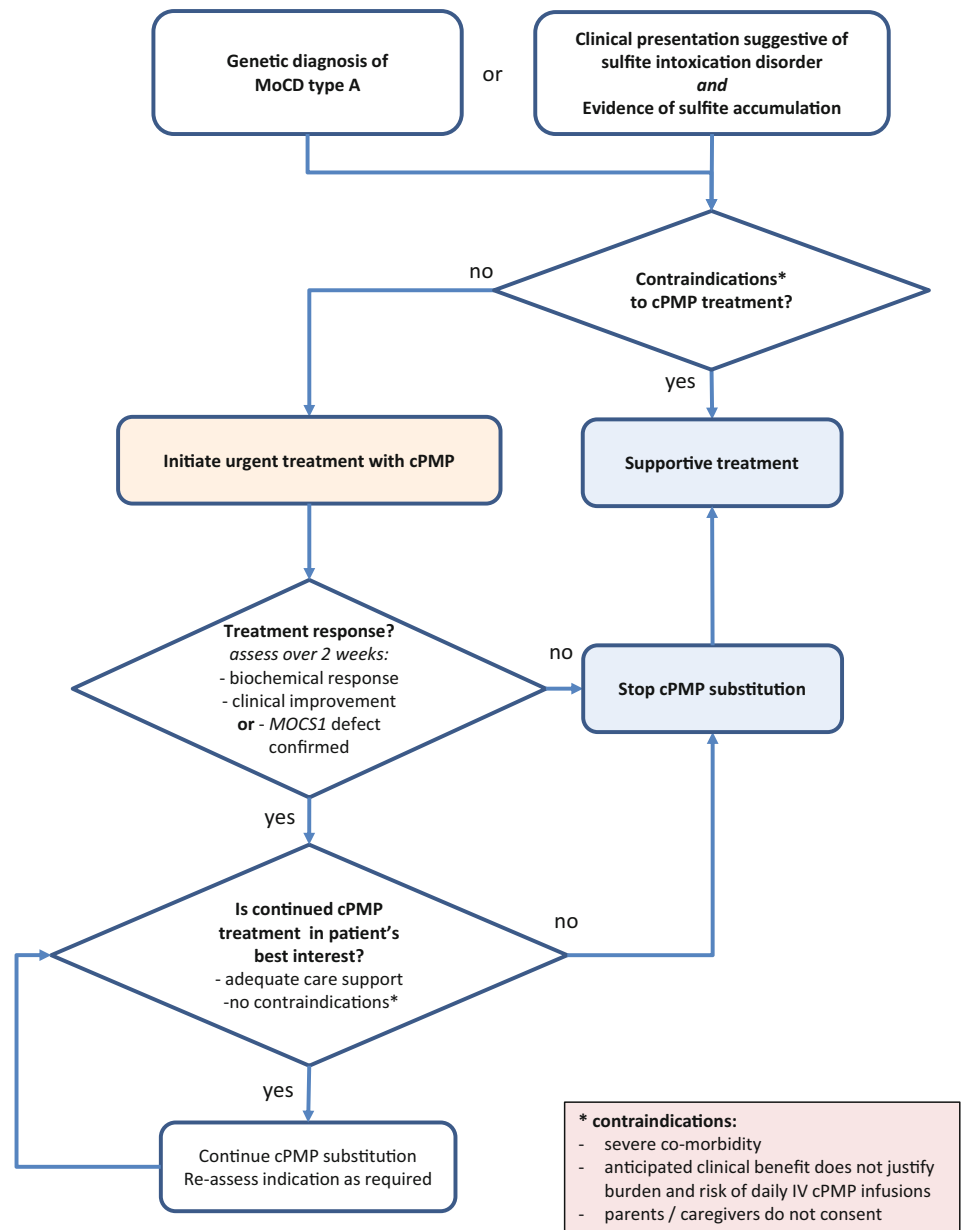
Results from a reduction of dietary sulfur intake with a diet low in methionine and cysteine have been reported in 4 patients with attenuated ISOD<sup>94,128,129</sup> and 7 patients with early onset typical disease<sup>47,130–135</sup>. Three out of 4 children with attenuated ISOD were deemed to benefit by showing decreased irritability and improved developmental progress<sup>94,128,129</sup> whereas 3 out of 7 children with typical ISOD were reported to benefit clinically with decreased irritability.<sup>130–132</sup>

Outcomes of a low sulfur diet were reported in 2 patients with attenuated MoCD<sup>15,136</sup> and in 5 patients with early onset typical MoCD.<sup>54,137–139</sup> The 2 children with attenuated MoCD and 1 out of 5 with typical MoCD<sup>137</sup> were reported to benefit with decreased irritability and improved developmental progress. Of note, one of the children with attenuated MoCD received a diet low in methionine but supplemented with cysteine.<sup>136</sup> The accumulation of sulfite-related biomarkers generally improved with dietary sulfur reduction.

#### Statement 33

Dietary sulfur restriction has been reported to provide clinical benefit for some patients with atypical presentations and attenuated disease. A clinical benefit for

**FIGURE 5** Therapy flowchart for suspected sulfite intoxication disorders.



patients with a typical presentation has not been consistently found.

A low-sulfur diet can be considered for children with atypical ISOD or MoCD and attenuated disease manifestations. Potential clinical benefits need to be balanced against poor palatability and requirements for supervision by a specialized dietitian and regular monitoring of plasma methionine concentrations to avoid protein malnutrition.

*Level of evidence: C*

*Strength of recommendation: Conditional*

*Expert opinion: 75% agree, 19% partly agree, 6% disagree (6/22 unable to comment)*

Statement 34

Dietary sulfur restriction is not required in children with MoCD-A who are treated with cPMP.

*Level of evidence: B*

*Strength of recommendation: Strong*

*Expert opinion: 87% agree, 13% partly agree (7/22 unable to comment)*

Good practice statement 35

If dietary treatment is deemed appropriate, sulfite accumulation can be decreased by reducing dietary sulfur intake to minimum requirements. This involves the restriction of natural protein to reduce the intake of the sulfur containing amino acids, methionine and cysteine. In published case studies the amount of methionine, from dietary protein, prescribed ranged from 18 to 30 mg/kg/day.<sup>94,129,130,137</sup>

Age	RNI for protein		Age	RNI for TSAA
	g/day	g/kg/day <sup>a</sup>		mg/kg/day
0–3 months	12.5 <sup>b</sup>	2.6	1 month	57
4–6 months	12.7	1.8	2 months	42
7–9 months	13.7	1.7	3 months	36
10–12 months	14.9	1.6	4–5 months	33
1–3 years	14.5	1.2	6–12 months	31
4–6 years	19.7		1–2 years	22
7–10 years	28.3		3–10 years	18
11–14 years (boys)	42.1			
11–14 years (girls)	41.2		11–14 years	17
15–18 years (boys)	55.5		15–18 years	16
15–18 years (girls)	45.4		>18 years	15

Note: Required nutritional intake (RNI) for protein<sup>140</sup> and for sulfur containing amino acids (TSAA).<sup>141</sup>

<sup>a</sup>Calculated using median weight from UK-WHO growth charts.

<sup>b</sup>No WHO figure given for infants aged 0–3 months. RNI calculated from recommendations of the UK Committee on Medical Aspects of Food Policy (COMA) [DH 1991].

Natural protein intake should be restricted to provide the minimum requirement for total sulfur containing amino acids (TSAA, methionine and cysteine) for age (Table 4). For individuals on liquid formula the volume of feed required to provide TSAA requirements can be calculated directly from the formula's stated methionine and cysteine content. If on a mixed diet information on amino acid contents of foods is less available. In this situation the methionine, or even protein content of the food may need to be used. Extrapolating from the adult TSAA requirement where individual methionine and cysteine requirements as well as TSAA are given, TSAA requirement is made of approximately 70% methionine and 30% cysteine. Thus to meet TSAA requirements for those on mixed diets 70% of the TSAA requirement can be calculated as a guide to methionine requirement.<sup>141</sup>

Overall total protein requirements should then be met using synthetic amino acid supplements without cysteine and methionine. It is suggested that total protein should equate to the minimum of RNI for age (Table 4) with an additional 20% to 40% to account for protein equivalence of amino acids being around 80% and the difference in amino acid absorption and metabolism compared to intact protein. Although there is no stipulation of protein content for amino acid based infant formulas, European Union regulations<sup>142</sup> specify that protein hydrolysate formula should contain 2.3–2.8 g protein/100 kcal compared with 1.8–2.5 g protein/100 kcal in standard cow milk based protein, suggesting that protein requirements from protein hydrolysate formula are higher than from standard cow milk protein based

TABLE 4 Nutritional requirements for a sulfur amino acid reduced diet.

formula. Based on this, it is likely that amino acid formulas would need to provide at a minimum the same protein as protein hydrolysate formula. Total energy requirements should meet standard energy requirements for age, for example Estimated Average Requirements,<sup>143</sup> which should then be adjusted for activity levels.

Dietary management should be supervised by a specialist metabolic dietitian. This would include regular reviews of growth and plasma quantitative amino acids including plasma methionine concentrations to avoid protein malnutrition. It is important that the clinical team and family review the efficacy of the diet on clinical symptoms such as seizures, neurological deterioration, and irritability to decide whether it is appropriate to continue.

Does molybdenum supplementation improve biochemical or neurological symptoms in MoCD?

A molybdenum supplement given to one child with early onset typical MoCD and one other with attenuated disease and did not provide any discernible biochemical or clinical improvement.<sup>136,138</sup> There is no plausible biological hypothesis to suggest efficacy of a molybdenum supplement and insufficient empirical evidence to determine the efficacy of a molybdenum supplement.

Statement 36

Molybdenum supplementation is not recommended for children with MoCD.

*Level of evidence: D*

*Strength of recommendation: Strong*

*Expert opinion: 94% agree, 6% partly agree (6/22 unable to comment)*

**Q10.** Is pyridoxine supplementation effective in ISOD and MoCD?

Sulfite chemically inactivates pyridoxal-5-phosphate (PLP), the active form of pyridoxine. Sulfite also strongly inhibits the enzyme alpha-AASA-dehydrogenase which leads to accumulation of alpha-aminoadipic semialdehyde (a-AASA) and delta-piperidine-6-carboxylate (P6C).<sup>74</sup> P6C inactivates PLP by condensation.<sup>74,144</sup> Both mechanisms contribute to functional PLP deficiency in ISOD and MoCD. A lack of PLP in CSF<sup>144</sup> and increased a-AASA in urine of patients with ISOD and MoCD<sup>45,74</sup> have been demonstrated. Functional PLP deficiency can contribute to seizures and disordered psychomotor development.<sup>145</sup>

Two siblings with neonatal typical MoCD-B were treated with pyridoxine (30 mg/kg) and folinic acid (3 mg/kg) on day 6 of life and at 2 years of age respectively. The neonatally treated girl remained seizure free at age 6 m, with feeding difficulties, microcephaly and hypotonia. The older sibling demonstrated a significant decrease in seizure frequency on the background of severe neurodisability when treatment was started.<sup>76</sup> Another patient with typical MoCD-B was treated at the age of 5 years with a pyridoxine supplement of 20 mg/kg per day and showed improvement in her energy levels and attentiveness.<sup>104</sup> Pyridoxine supplementation has been used in further typical cases of ISOD and MoCD with no obvious clinical benefit.<sup>92</sup>

**Statement 37**

There is clear evidence of functional PLP deficiency in ISOD or MoCD, but only limited evidence suggesting that pyridoxine supplementation is clinically effective in patients with typical ISOD or MoCD. Given the low risk and burden of the intervention we recommend to supplement pyridoxine to children diagnosed with ISOD or MoCD. The appropriate doses should be chosen in analogy to those used in pyridoxine dependent epilepsy.<sup>145</sup> Pyridoxine supplementation is not required in children with MoCD-A as long as they are treated with cPMP.

*Level of evidence: C*

*Strength of recommendation: Strong*

*Expert opinion: 89% agree, 11% partly agree (4/22 unable to comment)*

**Q11.** Is thiamine supplementation effective in ISOD and MoCD?

Thiamine is a water-soluble vitamin with rapid turnover and thiamine deficiency can develop over the course of a few weeks, leading to a disturbance in thiamine dependent mitochondrial energy metabolism and increased lactate concentrations. Sulfite readily inactivates thiamine

by nucleophilic cleavage of its intramolecular methylene bridge.<sup>146,147</sup> On this background it has been observed that patients with ISOD<sup>47,148,149</sup> and MoCD<sup>137,150,151</sup> can present with increased plasma lactate concentrations. This is often seen during the early neonatal encephalopathic phase of typical presentations. The hyperlactatemia is likely multifactorial and usually normalizes after a few weeks, but it has also been reported to persist for many months.<sup>137,149</sup> Hyperlactatemia is not always correlated with increased CSF lactate.<sup>148</sup> Thiamine supplementation has been attempted in MoCD<sup>138</sup> and ISOD<sup>47</sup> without discernible clinical benefit. However, long-term supplementation with thiamine has been recommended by some authors.<sup>69,107</sup>

**Statement 38**

There is no direct evidence to suggest that patients with ISOD and MoCD are experiencing functional thiamine deficiency. Thiamine supplementation may be considered in patients presenting with hyperlactatemia. A general recommendation for continued thiamine supplementation cannot be made on the background of current evidence.

*Level of evidence: D*

*Strength of recommendation: Conditional*

*Expert opinion: 100% agree, 0% partly agree, 0% disagree (4/22 unable to comment)*

## 5 | FAMILY PERSPECTIVE AND ADVOCACY ORGANIZATIONS

Patient/parent support and advocacy groups play an especially important role in ultra-rare diseases. They can be an invaluable source of information and of psychosocial support and can facilitate contact between carers of affected children. Due to the complex morbidity associated with sulfite intoxication disorders, patients are usually under the care of numerous medical specialties and require support from community-based healthcare and other professionals (e.g., education). Feedback from parents and support groups during the guideline development emphasized the importance of good communication between health professionals and families, as well as of access to resources, medical information and care support, which can be achieved by good signposting to relevant organizations.

The psychosocial impact on the whole family of having a child with a sulfite intoxication disorder cannot be underestimated. Families have commented that they value prioritization of comfort and quality of life when caring for a child with dystonic cerebral palsy and complex disability. They especially value access to neurophysiotherapy and specialist complex disability teams including for palliative care. A common theme has been

the experience of a general lack of knowledge and understanding for these rare conditions and issues arising from the delay in disease recognition and treatment. Various unmet needs were identified, including adequate symptom control, but also around support with tube feeding and with the long-term administration of daily enteral and intravenous medication.

### AUTHOR CONTRIBUTIONS

Bernd C. Schwahn, Francjan van Spronsen, Albert Misko, Ronen Spiegel, Guenter Schwarz, Flora Wong, Alistair Horman, James Pitt, Jörn Oliver Sass, and Charlotte Lubout led the systematic review and development of the initial statements. Julija Pavaine reviewed the neuroimaging sections, Victoria Holmes reviewed and amended the dietary section of the manuscript. All authors critically reviewed and contributed to the final manuscript and agreed to be accountable for the final version of the manuscript.

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### CONFLICT OF INTEREST STATEMENT

A competing interest statement is included in the Methods section of the manuscript.

### ETHICS STATEMENT

This article does not contain any studies with human or animal subjects performed by any of the authors.

Parents of affected children and the patient advocacy organization “Metabolic Support UK” consented to using their comments for this guideline. Additional specific consent was obtained for the use of photographs in Figure 2.

### ORCID

Bernd C. Schwahn  <https://orcid.org/0000-0003-0961-2508>

### REFERENCES

- Mudd SH, Irreverre F, Laster L. Sulfite oxidase deficiency in man: demonstration of the enzymatic defect. *Science*. 1967; 156(3782):1599-1602. doi:10.1126/science.156.3782.1599
- Johnson JL, Duran M, Sciver C, Beaudet A, Sly W, Valle D. Molybdenum cofactor deficiency and isolated sulfite oxidase deficiency. *The Metabolic and Molecular Bases of Inherited Disease*. McGraw-Hill; 2001.
- Schwarz G, Santamaria-Araujo JA, Wolf S, et al. Rescue of lethal molybdenum cofactor deficiency by a biosynthetic precursor from *Escherichia coli*. *Hum Mol Genet*. 2004;13(12): 1249-1255. doi:10.1093/hmg/ddh136
- Zhang X, Vincent AS, Halliwell B, Wong KP. A mechanism of sulfite neurotoxicity: direct inhibition of glutamate dehydrogenase. *J Biol Chem*. 2004;279(41):43035-43045. doi:10.1074/jbc.M402759200
- Grings M, Moura AP, Amaral AU, et al. Sulfite disrupts brain mitochondrial energy homeostasis and induces mitochondrial permeability transition pore opening via thiol group modification. *Biochim Biophys Acta*. 2014;1842(9):1413-1422. doi:10.1016/j.bbdis.2014.04.022
- Wyse ATS, Grings M, Wajner M, Leipnitz G. The role of oxidative stress and bioenergetic dysfunction in sulfite oxidase deficiency: insights from animal models. *Neurotox Res*. 2019; 35(2):484-494. doi:10.1007/s12640-018-9986-z
- Mellis AT, Roeper J, Misko AL, Kohl J, Schwarz G. Sulfite alters the mitochondrial network in molybdenum cofactor deficiency. *Front Genet*. 2021;11:594828. doi:10.3389/fgene.2020.594828
- Clarke H. The action of sulfite upon cystine. *J Biol Chem*. 1932;97:235-248.
- Olney JW, Misra CH, de Gubareff T. Cysteine-S-sulfate: brain damaging metabolite in sulfite oxidase deficiency. *J Neuropathol Exp Neurol*. 1975;34(2):167-177. doi:10.1097/00005072-197503000-00005
- Kumar A, Dejanovic B, Hetsch F, et al. S-sulfocysteine/NMDA receptor-dependent signaling underlies neurodegeneration in molybdenum cofactor deficiency. *J Clin Invest*. 2017;127(12):4365-4378. doi:10.1172/JCI89885
- Plate J, Sassen WA, Hassan AH, Lehne F, Köster RW, Kruse T. S-sulfocysteine induces seizure-like behaviors in zebrafish. *Front Pharmacol*. 2019;10:122. doi:10.3389/fphar.2019.00122
- Mayr S, May P, Arjune S, Havarushka N, Lal D, Schwarz G. Forecasting the incidence of rare diseases: an iterative computational and biochemical approach in molybdenum cofactor deficiency type a. *J Inherit Metab Dis*. 2019;42(S1):1-479. doi:10.1002/jimd.12153

13. Reiss J, Hahnwald R. Molybdenum cofactor deficiency: mutations in GPHN, MOCS1, and MOCS2. *Hum Mutat.* 2011; 32(1):10-18. doi:10.1002/humu.21390
14. Reiss J, Lenz U, Aquaviva-Bourdain C, Joriot-Chekaï S, Mention-Mulliez K, Holder-Espinasse M. A GPHN point mutation leading to molybdenum cofactor deficiency. *Clin Genet.* 2011;80(6):598-599. doi:10.1111/j.1399-0004.2011.01709.x
15. Huijman JGM, Schot R, de Klerk JBC, et al. Molybdenum cofactor deficiency: identification of a patient with homozygote mutation in the MOCS3 gene. *Am J Med Genet A.* 2017; 173(6):1601-1606. doi:10.1002/ajmg.a.38240
16. Schwahn B. Fosdenopterin: a first-in-class synthetic cyclic pyranopterin monophosphate for the treatment of molybdenum cofactor deficiency type A. *touchREVIEWS Neurol.* 2021; 17:85. Accessed February 22, 2022. <https://touchneurology.com/rare-diseases/journal-articles/fosdenopterin-a-first-in-class-synthetic-cyclic-pyranopterin-monophosphate-for-the-treatment-of-molybdenum-cofactor-deficiency-type-a/>
17. Guyatt GH, Oxman AD, Vist GE, et al. GRADE: an emerging consensus on rating quality of evidence and strength of recommendations. *BMJ.* 2008;336(7650):924-926. doi:10.1136/bmj.39489.470347.AD
18. Guyatt G, Oxman AD, Akl EA, et al. GRADE guidelines: 1. Introduction-GRADE evidence profiles and summary of findings tables. *J Clin Epidemiol.* 2011;64(4):383-394. doi:10.1016/j.jclinepi.2010.04.026
19. Balshem H, Helfand M, Schünemann HJ, et al. GRADE guidelines: 3. Rating the quality of evidence. *J Clin Epidemiol.* 2011;64(4):401-406. doi:10.1016/j.jclinepi.2010.07.015
20. Mechler K, Mountford WK, Hoffmann GF, Ries M. Ultra-orphan diseases: a quantitative analysis of the natural history of molybdenum cofactor deficiency. *Genet Med.* 2015;17(12):965-970. doi:10.1038/gim.2015.12
21. Claerhout H, Witters P, Régál L, et al. Isolated sulfite oxidase deficiency. *J Inher Metab Dis.* 2018;41(1):101-108. doi:10.1007/s10545-017-0089-4
22. Arican P, Gencpinar P, Kirbiyik O, et al. The clinical and molecular characteristics of molybdenum cofactor deficiency due to MOCS2 mutations. *Pediatr Neurol.* 2019;99:55-59. doi:10.1016/j.pediatrneurol.2019.04.021
23. Misko AL, Liang Y, Kohl JB, Eichler F. Delineating the phenotypic spectrum of sulfite oxidase and molybdenum cofactor deficiency. *Neurol Genet.* 2020;6(4):e486. doi:10.1212/NXG.0000000000000486
24. Spiegel R, Schwahn BC, Squires L, Confer N. Molybdenum cofactor deficiency: a natural history. *J Inher Metab Dis.* 2022; 45(3):456-469. doi:10.1002/jimd.12488
25. Schwahn BC, Van Spronsen FJ, Belaidi AA, et al. Efficacy and safety of cyclic pyranopterin monophosphate substitution in severe molybdenum cofactor deficiency type A: a prospective cohort study. *Lancet.* 2015;386(10007):1955-1963. doi:10.1016/S0140-6736(15)00124-5
26. Vijayakumar K, Gunny R, Grunewald S, et al. Clinical neuroimaging features and outcome in molybdenum cofactor deficiency. *Pediatr Neurol.* 2011;45(4):246-252. doi:10.1016/j.pediatrneurol.2011.06.006
27. Bayram E, Topcu Y, Karakaya P, et al. Molybdenum cofactor deficiency: review of 12 cases (MoCD and review). *Eur J Paediatr Neurol.* 2013;17(1):1-6. doi:10.1016/j.ejpn.2012.10.003
28. Zaki MS, Selim L, El-Bassyouni HT, et al. Molybdenum cofactor and isolated sulphite oxidase deficiencies: clinical and molecular spectrum among Egyptian patients. *Eur J Paediatr Neurol.* 2016;20(5):714-722. doi:10.1016/j.ejpn.2016.05.011
29. Macaya A, Brunso L, Fernández-Castillo N, et al. Molybdenum cofactor deficiency presenting as neonatal hyperekplexia: a clinical, biochemical and genetic study. *Neuropediatrics.* 2005;36(6):389-394. doi:10.1055/s-2005-872877
30. Lueder GT, Steiner RD. Ophthalmic abnormalities in molybdenum cofactor deficiency and isolated sulfite oxidase deficiency. *J Pediatr Ophthalmol Strabismus.* 1995;32(5):334-337.
31. De Klerk JB, Bakker HD, Beemer FA, et al. Facial dysmorphism in molybdenum cofactor deficiency/sulfite oxidase deficiency. *Enzyme Protein.* 1996;49:185.
32. Kaczmarek AT, Bender D, Gehling T, et al. A defect in molybdenum cofactor binding causes an attenuated form of sulfite oxidase deficiency. *J Inher Metab Dis.* 2022;45(2):169-182. doi:10.1002/jimd.12454
33. Hughes EF, Fairbanks L, Simmonds HA, Robinson RO. Molybdenum cofactor deficiency-phenotypic variability in a family with a late-onset variant. *Dev Med Child Neurol.* 1998; 40(1):57-61. doi:10.1111/j.1469-8749.1998.tb15357.x
34. Arenas M, Fairbanks LD, Vijayakumar K, Carr L, Escuredo E, Marinaki AM. An unusual genetic variant in the MOCS1 gene leads to complete missplicing of an alternatively spliced exon in a patient with molybdenum cofactor deficiency. *J Inher Metab Dis.* 2009;32(4):560-569. doi:10.1007/s10545-009-1151-7
35. Rocha S, Ferreira AC, Dias AI, Vieira JP, Sequeira S. Sulfite oxidase deficiency—an unusual late and mild presentation. *Brain Dev.* 2014;36(2):176-179. doi:10.1016/j.braindev.2013.01.013
36. Carman KB, Yildirim GK, Kiral E, et al. Status dystonicus: a rare presentation of molybdenum cofactor deficiency. *Int J Clin Pediatr.* 2017;6(3-4):51-53. doi:10.14740/ijcp283w
37. Mayr SJ, Sass JO, Vry J, et al. A mild case of molybdenum cofactor deficiency defines an alternative route of MOCS1 protein maturation. *J Inher Metab Dis.* 2018;41(2):187-196. doi:10.1007/s10545-018-0138-7
38. Scelsa B, Gasperini S, Righini A, Iacone M, Brazzoduro VG, Veggiotti P. Mild phenotype in molybdenum cofactor deficiency: a new patient and review of the literature. *Mol Genet Genomic Med.* 2019;7(6):e657. doi:10.1002/mgg3.657
39. Tian M, Qu Y, Huang L, et al. Stable clinical course in three siblings with late-onset isolated sulfite oxidase deficiency: a case series and literature review. *BMC Pediatr.* 2019;19(1):510. doi:10.1186/s12887-019-1889-5
40. Abe Y, Aihara Y, Endo W, et al. The effect of dietary protein restriction in a case of molybdenum cofactor deficiency with MOCS1 mutation. *Mol Genet Metab Rep.* 2021;26:100716. doi:10.1016/j.ymgmr.2021.100716
41. Tian Q, Cao Y, Shu L, et al. Case report: compound heterozygous variants in MOCS3 identified in a Chinese infant with molybdenum cofactor deficiency. *Front Genet.* 2021;12:651878. doi:10.3389/fgene.2021.651878
42. Stiburkova B. Mild phenotype and late onset in molybdenum cofactor deficiency: a novel variant in MOCS2 gene. In: Roma Population Poster B83. SSIEM Annual Symposium 2023. Published online August 29, 2023.

43. Sharawat IK, Saini L, Singanamala B, et al. Metabolic crisis after trivial head trauma in late-onset isolated sulfite oxidase deficiency: report of two new cases and review of published patients. *Brain Dev.* 2020;42(2):157-164. doi:10.1016/j.braindev.2019.11.003
44. Schwarz G, Mendel RR, Ribbe MW. Molybdenum cofactors, enzymes and pathways. *Nature.* 2009;460(7257):839-847. doi:10.1038/nature08302
45. Kožich V, Schwahn BC, Sokolová J, et al. Human ultrarare genetic disorders of sulfur metabolism demonstrate redundancies in H<sub>2</sub>S homeostasis. *Redox Biol.* 2022;58:102517. doi:10.1016/j.redox.2022.102517
46. Mraz M, Hurba O, Bartl J, et al. Modern diagnostic approach to hereditary xanthinuria. *Urolithiasis.* 2015;43(1):61-67. doi:10.1007/s00240-014-0734-4
47. Tan WH, Eichler FS, Hoda S, et al. Isolated sulfite oxidase deficiency: a case report with a novel mutation and review of the literature. *Pediatrics.* 2005;116(3):757-766. doi:10.1542/peds.2004-1897
48. Simmonds HA, Hoffmann GF, Pérignon JL, Micheli V, van Gennip AH. Diagnosis of molybdenum cofactor deficiency. *Lancet.* 1999;353(9153):675. doi:10.1016/s0140-6736(05)75473-8
49. van der Klei-van Moorsel JM, Smit LM, Brockstedt M, Jakobs C, Dorche C, Duran M. Infantile isolated sulphite oxidase deficiency: report of a case with negative sulphite test and normal sulphate excretion. *Eur J Pediatr.* 1991;150(3):196-197. doi:10.1007/BF01963565
50. Duran M, Aarsen G, Fokkens RH, et al. 2-Mercaptoethanesulfonate-cysteine disulfide excretion following the administration of 2-mercaptoethanesulfonate—a pitfall in the diagnosis of sulfite oxidase deficiency. *Clin Chim Acta.* 1981;111(1):47-53. doi:10.1016/0009-8981(81)90419-8
51. Wadman SK, Duran M, Beemer FA, et al. Absence of hepatic molybdenum cofactor: an inborn error of metabolism leading to a combined deficiency of sulphite oxidase and xanthine dehydrogenase. *J Inherit Metab Dis.* 1983;6(Suppl 1):78-83. doi:10.1007/BF01811328
52. Irreverre F, Mudd SH, Heizer WD, Laster L. Sulfite oxidase deficiency: studies of a patient with mental retardation, dislocated ocular lenses, and abnormal urinary excretion of S-sulfo-l-cysteine, sulfite, and thiosulfate. *Biochem Med.* 1967;1(2):187-217. doi:10.1016/0006-2944(67)90007-5
53. van Gennip AH, Abeling NG, Stroomeer AE, Overmars H, Bakker HD. The detection of molybdenum cofactor deficiency: clinical symptomatology and urinary metabolite profile. *J Inherit Metab Dis.* 1994;17(1):142-145. doi:10.1007/BF00735420
54. Duran M, Beemer FA, van de Heiden C, et al. Combined deficiency of xanthine oxidase and sulphite oxidase: a defect of molybdenum metabolism or transport? *J Inherit Metab Dis.* 1978;1(4):175-178. doi:10.1007/BF01805591
55. Belaidi AA, Arjune S, Santamaria-Araujo JA, Sass JO, Schwarz G. Molybdenum cofactor deficiency: a new HPLC method for fast quantification of s-sulfocysteine in urine and serum. *JIMD Rep.* 2012;5:35-43. doi:10.1007/8904\_2011\_89
56. Rashed MS, Saadallah AAA, Rahbeeni Z, et al. Determination of urinary S-sulphocysteine, xanthine and hypoxanthine by liquid chromatography-electrospray tandem mass spectrometry. *Biomed Chromatogr.* 2005;19(3):223-230. doi:10.1002/bmc.439
57. Pitt JJ, Eggington M, Kahler SG. Comprehensive screening of urine samples for inborn errors of metabolism by electrospray tandem mass spectrometry. *Clin Chem.* 2002;48(11):1970-1980.
58. Belaidi AA, Schwarz G. Molybdenum cofactor deficiency: metabolic link between taurine and S-sulfocysteine. *Adv Exp Med Biol.* 2013;776:13-19. doi:10.1007/978-1-4614-6093-0\_2
59. Bremer HJ, Duran M, Kamerling JP, Przyrembel H, Wadman SK. *Disturbances of Amino Acid Metabolism: Clinical Chemistry and Diagnosis.* Urban & Schwarzenberg; 1981.
60. Choong CT, Hart C, Tan ES, Goh DLM. Clinical and laboratory barriers to the timely diagnosis of Sulphite oxidase deficiency. *Proc Singap Healthc.* 2010;19(2):94-100. doi:10.1177/201010581001900203
61. Shih VE, Carney MM, Mandell R. A simple screening test for sulfite oxidase deficiency: detection of urinary thiosulfate by a modification of Sörbo's method. *Clin Chim Acta.* 1979;95(1):143-145. doi:10.1016/0009-8981(79)90348-6
62. Shih VE. Ampicillin interference with test for sulfite oxidase deficiency. *Clin Chem.* 1983;29(1):211-212.
63. Mann G, Kirk JM. Antibiotic interference in urinary thiosulphate measurements. *J Inherit Metab Dis.* 1994;17(1):120-121. doi:10.1007/BF00735412
64. Carragher FM, Kirk JM, Steer C, Allen J, Dorche C. False negative thiosulphate screening test in a case of molybdenum cofactor deficiency. *J Inherit Metab Dis.* 1999;22(7):842-843. doi:10.1023/a:1005518426781
65. Veldman A, Santamaria-Araujo JA, Sollazzo S, et al. Successful treatment of molybdenum cofactor deficiency type A with cPMP. *Pediatrics.* 2010;125(5):e1249-e1254. doi:10.1542/peds.2009-2192
66. Graf WD, Oleinik OE, Jack RM, Weiss AH, Johnson JL. Ahomocysteinemia in molybdenum cofactor deficiency. *Neurology.* 1998;51(3):860-862. doi:10.1212/wnl.51.3.860
67. Sass JO, Nakanishi T, Sato T, Shimizu A. New approaches towards laboratory diagnosis of isolated sulphite oxidase deficiency. *Ann Clin Biochem.* 2004;41(Pt 2):157-159. doi:10.1258/000456304322880078
68. Kaczmarek AT, Bahlmann N, Thaqi B, May P, Schwarz G. Machine learning-based identification and characterization of 15 novel pathogenic SUOX missense mutations. *Mol Genet Metab.* 2021;134(1-2):188-194. doi:10.1016/j.ymgme.2021.07.011
69. Bindu PS, Nagappa M, Bharath RD, Taly AB. Isolated sulfite oxidase deficiency. In: Adam MP, Mirzaa GM, Pagon RA, et al., eds. *GeneReviews*®. University of Washington; 1993. Accessed June 3, 2022. <http://www.ncbi.nlm.nih.gov/books/NBK453433/>
70. Lee HF, Chi CS, Tsai CR, Chen HC, Lee IC. Prenatal brain disruption in isolated sulfite oxidase deficiency. *Orphanet J Rare Dis.* 2017;12(1):115. doi:10.1186/s13023-017-0668-3
71. Haschke-Becher E, Kainz A, Bachmann C. Reference values of amino acids and of common clinical chemistry in plasma of healthy infants aged 1 and 4 months. *J Inherit Metab Dis.* 2016;39(1):25-37. doi:10.1007/s10545-015-9870-4
72. Sebesta I, Stiburkova B, Krijt J. Hereditary xanthinuria is not so rare disorder of purine metabolism. *Nucleosides Nucleotides*

- Nucleic Acids*. 2018;37(6):324-328. doi:[10.1080/15257770.2018.1460478](https://doi.org/10.1080/15257770.2018.1460478)
73. Castro M, Carrillo R, García F, et al. Thirteen years experience with selective screening for disorders in purine and pyrimidine metabolism. *Nucleosides Nucleotides Nucleic Acids*. 2014; 33(4-6):233-240. doi:[10.1080/15257770.2013.854381](https://doi.org/10.1080/15257770.2013.854381)
  74. Mills PB, Footitt EJ, Ceyhan S, et al. Urinary AASA excretion is elevated in patients with molybdenum cofactor deficiency and isolated sulphite oxidase deficiency. *J Inherit Metab Dis*. 2012;35(6):1031-1036. doi:[10.1007/s10545-012-9466-1](https://doi.org/10.1007/s10545-012-9466-1)
  75. Carmi-Nawi N, Malinger G, Mandel H, Ichida K, Lerman-Sagie T, Lev D. Prenatal brain disruption in molybdenum cofactor deficiency. *J Child Neurol*. 2011;26(4):460-464. doi:[10.1177/0883073810383017](https://doi.org/10.1177/0883073810383017)
  76. Struys EA, Nota B, Bakkali A, Al Shahwan S, Salomons GS, Tabarki B. Pyridoxine-dependent epilepsy with elevated urinary  $\alpha$ -amino adipic semialdehyde in molybdenum cofactor deficiency. *Pediatrics*. 2012;130(6):e1716-e1719. doi:[10.1542/peds.2012-1094](https://doi.org/10.1542/peds.2012-1094)
  77. Kishikawa M, Sass JO, Sakura N, Nakanishi T, Shimizu A, Yoshioka M. The peak height ratio of S-sulfonated transthyretin and other oxidized isoforms as a marker for molybdenum cofactor deficiency, measured by electrospray ionization mass spectrometry. *Biochim Biophys Acta*. 2002;1588(2):135-138. doi:[10.1016/s0925-4439\(02\)00156-4](https://doi.org/10.1016/s0925-4439(02)00156-4)
  78. Johnson JL, Rajagopalan KV. Structural and metabolic relationship between the molybdenum cofactor and urothione. *Proc Natl Acad Sci U S A*. 1982;79(22):6856-6860. doi:[10.1073/pnas.79.22.6856](https://doi.org/10.1073/pnas.79.22.6856)
  79. Pristup J, Schaeffeler E, Arjune S, et al. Molybdenum cofactor catabolism unravels the physiological role of the drug metabolizing enzyme thiopurine S-methyltransferase. *Clin Pharmacol Ther*. 2022;112(4):808-816. doi:[10.1002/cpt.2637](https://doi.org/10.1002/cpt.2637)
  80. Hitzert MM, Bos AF, Bergman KA, et al. Favorable outcome in a newborn with molybdenum cofactor type A deficiency treated with cPMP. *Pediatrics*. 2012;130(4):e1005-e1010. doi:[10.1542/peds.2011-3330](https://doi.org/10.1542/peds.2011-3330)
  81. Westerlinck H, Meylaerts L, Van Hoestenbergh MR, Rossi A. Sulfite oxidase deficiency in a newborn. *JBR-BTR*. 2014;97(2): 113-114. doi:[10.5334/jbr-btr.40](https://doi.org/10.5334/jbr-btr.40)
  82. Hannah-Shmouni F, MacNeil L, Potter M, et al. Severe cystic degeneration and intractable seizures in a newborn with molybdenum cofactor deficiency type B. *Mol Genet Metab Rep*. 2019;18:11-13. doi:[10.1016/j.jymgmr.2018.12.003](https://doi.org/10.1016/j.jymgmr.2018.12.003)
  83. Alonzo Martínez MC, Cazorla E, Cánovas E, Anniuk K, Cores AE, Serrano AM. Molybdenum cofactor deficiency: mega cisterna magna in two consecutive pregnancies and review of the literature. *Appl Clin Genet*. 2020;13:49-55. doi:[10.2147/TACG.S239917](https://doi.org/10.2147/TACG.S239917)
  84. Lubout CMA, Derks TGJ, Meiners L, et al. Molybdenum cofactor deficiency type A: prenatal monitoring using MRI. *Eur J Paediatr Neurol*. 2018;22(3):536-540. doi:[10.1016/j.ejpn.2017.11.006](https://doi.org/10.1016/j.ejpn.2017.11.006)
  85. Chen LW, Tsai YS, Huang CC. Prenatal multicystic encephalopathy in isolated sulfite oxidase deficiency with a novel mutation. *Pediatr Neurol*. 2014;51(1):181-182. doi:[10.1016/j.pediatrneurol.2014.03.010](https://doi.org/10.1016/j.pediatrneurol.2014.03.010)
  86. Stence NV, Coughlin CR, Fenton LZ, Thomas JA. Distinctive pattern of restricted diffusion in a neonate with molybdenum cofactor deficiency. *Pediatr Radiol*. 2013;43(7):882-885. doi:[10.1007/s00247-012-2579-8](https://doi.org/10.1007/s00247-012-2579-8)
  87. Higuchi R, Sugimoto T, Tamura A, et al. Early features in neuroimaging of two siblings with molybdenum cofactor deficiency. *Pediatrics*. 2014;133(1):e267-e271. doi:[10.1542/peds.2013-0935](https://doi.org/10.1542/peds.2013-0935)
  88. Lin Y, Liu Y, Chen S, et al. A neonate with molybdenum cofactor deficiency type B. *Transl Pediatr*. 2021;10(4):1039-1044. doi:[10.21037/tp-20-357](https://doi.org/10.21037/tp-20-357)
  89. Li JT, Chen ZX, Chen XJ, Jiang YX. Mutation analysis of SUOX in isolated sulfite oxidase deficiency with ectopia lentis as the presenting feature: insights into genotype-phenotype correlation. *Orphanet J Rare Dis*. 2022;17(1):392. doi:[10.1186/s13023-022-02544-x](https://doi.org/10.1186/s13023-022-02544-x)
  90. Hobson EE, Thomas S, Crofton PM, Murray AD, Dean JCS, Lloyd D. Isolated sulphite oxidase deficiency mimics the features of hypoxic ischaemic encephalopathy. *Eur J Pediatr*. 2005;164(11):655-659. doi:[10.1007/s00431-005-1729-5](https://doi.org/10.1007/s00431-005-1729-5)
  91. Eyaid WM, Al-Nouri DM, Rashed MS, Al-Rifai MT, Al-Wakeel AS. An inborn error of metabolism presenting as hypoxic-ischemic insult. *Pediatr Neurol*. 2005;32(2):134-136. doi:[10.1016/j.pediatrneurol.2004.07.010](https://doi.org/10.1016/j.pediatrneurol.2004.07.010)
  92. Own Observation.
  93. Yoganathan S, Sudhakar S, Thomas M, Kumar Dutta A, Danda S, Chandran M. Novel imaging finding and novel mutation in an infant with molybdenum cofactor deficiency, a mimicker of hypoxic-Ischaemic encephalopathy. *Iran J Child Neurol*. 2018;12(2):107-112.
  94. Del Rizzo M, Burlina AP, Sass JO, et al. Metabolic stroke in a late-onset form of isolated sulfite oxidase deficiency. *Mol Genet Metab*. 2013;108(4):263-266. doi:[10.1016/j.ymgme.2013.01.011](https://doi.org/10.1016/j.ymgme.2013.01.011)
  95. Alkufri F, Harrower T, Rahman Y, et al. Molybdenum cofactor deficiency presenting with a parkinsonism-dystonia syndrome. *Mov Disord*. 2013;28(3):399-401. doi:[10.1002/mds.25276](https://doi.org/10.1002/mds.25276)
  96. Pavel AM, Stephens CM, Mathieson SR, et al. Case report: electroencephalography in a neonate with isolated sulfite oxidase deficiency—a case report and literature review. *HRB Open Res*. 2021;4:122. doi:[10.12688/hrbopenres.13442.1](https://doi.org/10.12688/hrbopenres.13442.1)
  97. Megahed H, Nicouveau M, Barcia G, et al. Utility of whole exome sequencing for the early diagnosis of pediatric-onset cerebellar atrophy associated with developmental delay in an inbred population. *Orphanet J Rare Dis*. 2016;11(1):57. doi:[10.1186/s13023-016-0436-9](https://doi.org/10.1186/s13023-016-0436-9)
  98. Botkin JR, Rothwell E. Whole genome sequencing and newborn screening. *Curr Genet Med Rep*. 2016;4(1):1-6. doi:[10.1007/s40142-016-0084-3](https://doi.org/10.1007/s40142-016-0084-3)
  99. Jasinge E, Kularatnam GAM, Dilanthi HW, et al. Uric acid, an important screening tool to detect inborn errors of metabolism: a case series. *BMC Res Notes*. 2017;10(1):454. doi:[10.1186/s13104-017-2795-2](https://doi.org/10.1186/s13104-017-2795-2)
  100. Currie S, Claire G. A Case of Isolated Sulfite Oxidase Deficiency and Pyloric Stenosis: Coincidence or Common Etiology? *J Pediatr Neurol*. 2009;7:181-186. doi:[10.3233/JPN-2009-0280](https://doi.org/10.3233/JPN-2009-0280)
  101. Tezel G, Oztekin O, Kalay S, Aslan A, Akçağuş M, Oygür N. The association of molybdenum cofactor deficiency and pyloric stenosis. *J Perinatol*. 2012;32(11):896-898. doi:[10.1038/jp.2011.192](https://doi.org/10.1038/jp.2011.192)



102. Satar M, Kurtoğlu Aİ, Yıldızdaş HY, Önenli Mungan N, Özlü F, Bişgin A. Coexistence of molybdenum cofactor deficiency type A and hypertrophic pyloric stenosis, a new case. *Turk Arch Pediatr.* 2021;56(1):78-80. doi:10.14744/TurkPediatriArs.2020.57984
103. Karunakar P, Krishnamurthy S, Kasinathan A, Hariharan R, Chidambaram AC. Renal stones in an infant with microcephaly and spastic quadriplegia: answers. *Pediatr Nephrol.* 2020;35(6):987-989. doi:10.1007/s00467-019-04449-7
104. Lee EJ, Dandamudi R, Granadillo JL, Grange DK, Kakajiwala A. Rare cause of xanthinuria: a pediatric case of molybdenum cofactor deficiency B. *CEN Case Rep.* 2021;10(3):378-382. doi:10.1007/s13730-021-00572-3
105. Hinderhofer K, Mechler K, Hoffmann GF, Lampert A, Mountford WK, Ries M. Critical appraisal of genotype assessment in molybdenum cofactor deficiency. *J Inherit Metab Dis.* 2017;40(6):801-811. doi:10.1007/s10545-017-0077-8
106. Bender D, Kaczmarek AT, Kuester S, Burlina AB, Schwarz G. Oxygen and nitrite reduction by heme-deficient sulphite oxidase in a patient with mild sulphite oxidase deficiency. *J Inherit Metab Dis.* 2020;43(4):748-757. doi:10.1002/jimd.12216
107. Misko A, Mahtani K, Abbott J, Schwarz G, Atwal P. Molybdenum cofactor deficiency. In: Adam MP, Ardinger HH, Pagon RA, et al., eds. *GeneReviews*<sup>®</sup>. University of Washington; 1993. Accessed March 17, 2022. <http://www.ncbi.nlm.nih.gov/books/NBK575630/>
108. Johannes L, Fu CY, Schwarz G. Molybdenum cofactor deficiency in humans. *Molecules.* 2022;27(20):6896. doi:10.3390/molecules27206896
109. Veldman A, Hennermann JB, Schwarz G, et al. Timing of cerebral developmental disruption in molybdenum cofactor deficiency. *J Child Neurol.* 2011;26(8):1059-1060; author reply 1061. doi:10.1177/0883073811415851
110. Fatemi A, Wilson MA, Johnston MV. Hypoxic-ischemic encephalopathy in the term infant. *Clin Perinatol.* 2009;36(4):835-858, vii. doi:10.1016/j.clp.2009.07.011
111. Schuierer G, Kurlmann G, Bick U, Stephani U. Molybdenum-cofactor deficiency: CT and MR findings. *Neuropediatrics.* 1995;26(1):51-54. doi:10.1055/s-2007-979720
112. Schwahn BC. Postnatal hepatocyte transplant in MoCD type B Poster 596. SSIEM 2016 Annual Symposium—Abstracts. *J Inherit Metab Dis.* 2016;39:35-284. doi:10.1007/s10545-016-9969-2
113. Chen X, Malaeb SN, Pan J, Wang L, Scafidi J. Editorial: perinatal hypoxic-ischemic brain injury: mechanisms, pathogenesis, and potential therapeutic strategies. *Front Cell Neurosci.* 2022;16:1086692. doi:10.3389/fncel.2022.1086692
114. Pena AB, Caviness JN. Physiology-based treatment of myoclonus. *Neurotherapeutics.* 2020;17(4):1665-1680. doi:10.1007/s13311-020-00922-6
115. Albanese A, Bhatia K, Bressman SB, et al. Phenomenology and classification of dystonia: a consensus update. *Mov Disord.* 2013;28(7):863-873. doi:10.1002/mds.25475
116. Lumsden DE, Gimeno H, Lin JP. Classification of dystonia in childhood. *Parkinsonism Relat Disord.* 2016;33:138-141. doi:10.1016/j.parkreldis.2016.10.001
117. Yoshimura A, Kibe T, Hasegawa H, et al. The persistent generalized muscle contraction in siblings with molybdenum cofactor deficiency type a. *Neuropediatrics.* 2019;50(2):126-129. doi:10.1055/s-0039-1677869
118. Monbaliu E. Clinical presentation and management of dyskinetic cerebral palsy. *Lancet Neurol.* 2017;16:741-749.
119. Harvey A, Reddihough D, Scheinberg A, Williams K. Oral medication prescription practices of tertiary-based specialists for dystonia in children with cerebral palsy. *J Paediatr Child Health.* 2018;54(4):401-404. doi:10.1111/jpc.13754
120. Lumsden DE, King MD, Allen NM. Status dystonicus in childhood. *Curr Opin Pediatr.* 2017;29(6):674-682. doi:10.1097/MOP.0000000000000556
121. Yan W, Huang L, Sun L, Ding X. Ocular characteristics of a 6-year-old boy with molybdenum cofactor deficiency type B. *Am J Ophthalmol Case Rep.* 2022;27:101586. doi:10.1016/j.ajoc.2022.101586
122. Arikyants N, Sarkissian A, Hesse A, Eggermann T, Leumann E, Steinmann B. Xanthinuria type I: a rare cause of urolithiasis. *Pediatr Nephrol.* 2007;22(2):310-314. doi:10.1007/s00467-006-0267-3
123. Cartier P, Perignon JL. [Xanthinuria (author's transl)]. *Nouv Presse Med.* 1978;7(16):1381-1390.
124. Prescribing information for fosdenopterin hydrobromide dihydrate (NULIBRY TM). Accessed December 27, 2023. <https://dailymed.nlm.nih.gov/dailymed/drugInfo.cfm?setid=736aeea3-f206-454d-95d0-fd30964e8a8a> or [https://www.ema.europa.eu/en/documents/product-information/nulibry-epar-product-information\\_en.pdf](https://www.ema.europa.eu/en/documents/product-information/nulibry-epar-product-information_en.pdf)
125. Kang C. Fosdenopterin: first approval. *Drugs.* 2021;81(8):953-956. doi:10.1007/s40265-021-01520-2
126. Watsky E, Kuklin N, Hamilton M, Barr S. Maintenance of disease-relevant biomarker improvement in patients with molybdenum cofactor deficiency (MoCD) type A administered ALXN1101, a synthetic form of cyclic pyranopterin monophosphate. Presented at 12th EPNS Congress, Lyon France 22 June 2017, pp. 2-87. JPN. Published online 2017. 21(S1):E124.
127. Farrell S, Karp J, Hager R, et al. Regulatory news: Nulibry (fosdenopterin) approved to reduce the risk of mortality in patients with molybdenum cofactor deficiency type a: FDA approval summary. *J Inherit Metab Dis.* 2021;44(5):1085-1087. doi:10.1002/jimd.12421
128. Shih VE, Abroms IF, Johnson JL, et al. Sulfite oxidase deficiency. Biochemical and clinical investigations of a hereditary metabolic disorder in sulfur metabolism. *N Engl J Med.* 1977;297(19):1022-1028. doi:10.1056/NEJM197711102971902
129. Touati G, Rusthoven E, Depondt E, et al. Dietary therapy in two patients with a mild form of sulphite oxidase deficiency. Evidence for clinical and biological improvement. *J Inherit Metab Dis.* 2000;23(1):45-53. doi:10.1023/a:1005646813492
130. Boyer M, Sowa M, Wang R, Abdenur J. Isolated sulfite oxidase deficiency: response to dietary treatment in a patient with severe neonatal presentation. *J Inborn Errors Metab Screen.* 2019;7:e20190001. doi:10.1590/2326-4594-jiems-2019-0001
131. Chan KY, Li CK, Lai CK, Ng SF, Chan AYW. Infantile isolated sulphite oxidase deficiency in a Chinese family: a rare neurodegenerative disorder. *Hong Kong Med J.* 2002;8(4):279-282.
132. Lee HJ, Adham IM, Schwarz G, et al. Molybdenum cofactor-deficient mice resemble the phenotype of human patients.

- Hum Mol Genet.* 2002;11(26):3309-3317. doi:10.1093/hmg/11.26.3309
133. Dublin AB, Hald JK, Wootton-Gorges SL. Isolated sulfite oxidase deficiency: MR imaging features. *AJNR Am J Neuroradiol.* 2002;23(3):484-485.
  134. Sass JO, Gunduz A, Araujo Rodrigues Funayama C, et al. Functional deficiencies of sulfite oxidase: differential diagnoses in neonates presenting with intractable seizures and cystic encephalomalacia. *Brain Dev.* 2010;32(7):544-549. doi:10.1016/j.braindev.2009.09.005
  135. Holder JL, Agadi S, Reese W, Rehder C, Quach MM. Infantile spasms and hyperekplexia associated with isolated sulfite oxidase deficiency. *JAMA Neurol.* 2014;71(6):782-784. doi:10.1001/jamaneurol.2013.5083
  136. Bamforth FJ, Johnson JL, Davidson AG, Wong LT, Lockitch G, Applegarth DA. Biochemical investigation of a child with molybdenum cofactor deficiency. *Clin Biochem.* 1990;23(6):537-542. doi:10.1016/0009-9120(90)80046-1
  137. Boles RG, Ment LR, Meyn MS, Horwich AL, Kratz LE, Rinaldo P. Short-term response to dietary therapy in molybdenum cofactor deficiency. *Ann Neurol.* 1993;34(5):742-744. doi:10.1002/ana.410340520
  138. Endres W, Shin YS, Günther R, Ibel H, Duran M, Wadman SK. Report on a new patient with combined deficiencies of sulphite oxidase and xanthine dehydrogenase due to molybdenum cofactor deficiency. *Eur J Pediatr.* 1988;148(3):246-249. doi:10.1007/BF00441412
  139. Ngu LH, Afroze B, Chen BC, Affandi O, Zabedah MY. Molybdenum cofactor deficiency in a Malaysian child. *Singapore Med J.* 2009;50(10):e365-e367.
  140. Dietary reference values for food energy and nutrients for the United Kingdom. Report of the panel on dietary reference values of the committee on medical aspects of food policy. *Rep Health Soc Subj (Lond).* 1991;41:1-210.
  141. Joint WHO/FAO/UNU Expert Consultation. Protein and amino acid requirements in human nutrition. *World Health Organ Tech Rep Ser.* 2007;(935):1-265. Back cover. PMID: 18330140.
  142. Commission Delegated Regulation (EU) 2022/519 of 14 January 2022 amending Delegated Regulation (EU) 2016/127 as regards the protein requirements for infant and follow-on formula manufactured from protein hydrolysates (Text with EEA relevance). Document 32022R0519. ELI. Accessed December 27, 2023. [http://data.europa.eu/eli/reg\\_del/2022/519/oj](http://data.europa.eu/eli/reg_del/2022/519/oj)
  143. Dietary Reference Values for Energy. *Scientific Advisory Committee on Nutrition.* 2011. Accessed December 27, 2023. <https://www.gov.uk/government/publications/sacn-dietary-reference-values-for-energy>
  144. Footitt EJ, Heales SJ, Mills PB, Allen GFG, Oppenheim M, Clayton PT. Pyridoxal 5'-phosphate in cerebrospinal fluid; factors affecting concentration. *J Inherit Metab Dis.* 2011;34(2):529-538. doi:10.1007/s10545-011-9279-7
  145. Coughlin CR, Tseng LA, Abdenur JE, et al. Consensus guidelines for the diagnosis and management of pyridoxine-dependent epilepsy due to  $\alpha$ -aminoadipic semialdehyde dehydrogenase deficiency. *J Inherit Metab Dis.* 2021;44(1):178-192. doi:10.1002/jimd.12332
  146. Williams RR, Waterman RE, Keresztesy JC, Buchman ER. Studies of crystalline vitamin B1. III. Cleavage of vitamin with sulfite. *J Am Chem Soc.* 1935;57(3):536-537. doi:10.1021/ja01306a044
  147. Zoltewicz J, Kauffman G, Uray G. A Mechanism for Sulphite Ion Reacting With Vitamin B1 and Its Analogues. *Food Chem.* 1984;15:75-91.
  148. Rupar CA, Gillett J, Gordon BA, et al. Isolated sulfite oxidase deficiency. *Neuropediatrics.* 1996;27(6):299-304. doi:10.1055/s-2007-973798
  149. Basheer SN, Waters PJ, Lam CW, et al. Isolated sulfite oxidase deficiency in the newborn: lactic acidemia and leukoencephalopathy. *Neuropediatrics.* 2007;38(1):38-41. doi:10.1055/s-2007-981484
  150. Arnold GL, Greene CL, Stout JP, Goodman SI. Molybdenum cofactor deficiency. *J Pediatr.* 1993;123(4):595-598. doi:10.1016/s0022-3476(05)80961-8
  151. Appignani BA, Kaye EM, Wolpert SM. CT and MR appearance of the brain in two children with molybdenum cofactor deficiency. *AJNR Am J Neuroradiol.* 1996;17(2):317-320.

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